22 September 2006 | $10

Science

Visualization Challenge 2006

AAAS

Volume 313, Issue 5794

COVER

A computer-generated rendering of five mathematical surfaces, depicted as glassy objects on a glass tabletop. This image was awarded first place in the illustration category of the National Science Foundation/*Science* 2006 Visualization Challenge. All of the winning entries are described in a special feature beginning on page 1729.

Illustration: R. Palais and L. Benard

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Rapid Chemically Induced Changes of PtdIns(4,5)P_2 Gate KCNQ Ion Channels

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Neurotransmitters close a potassium channel by changing the lipid content of the surrounding plasma membrane.

10.1126/science.1131163

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M. Willem et al.

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10.1126/science.1132341

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10.1126/science.1131794

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Response to Comments on "A Keystone Mutualism Drives Pattern in a Power Function"

J. Vandermeer and I. Perfecto

full text at www.sciencemag.org/cgi/content/full/313/5794/1739d

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***SUMO1* Haploinsufficiency Leads to Cleft Lip and Palate**

F. S. Alkuraya et al.

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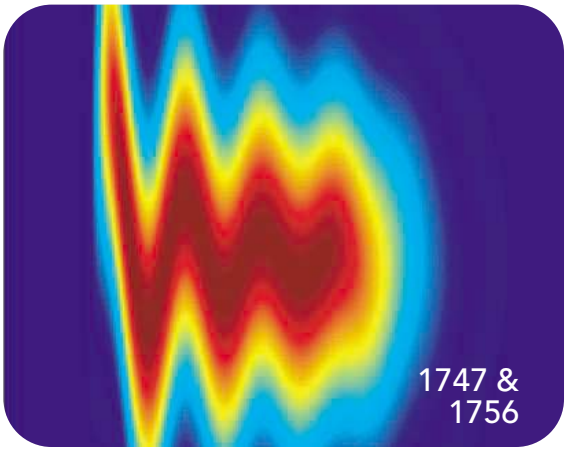

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Structural Basis of *glmS* Ribozyme Activation by Glucosamine-6-Phosphate

D. J. Klein and A. R. Ferré-D'Amaré

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P. R. Poulin and K. A. Nelson

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R. C. Greenwood et al.

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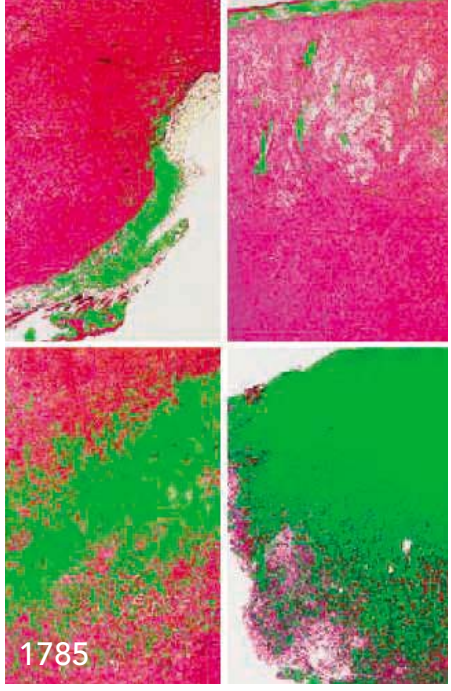

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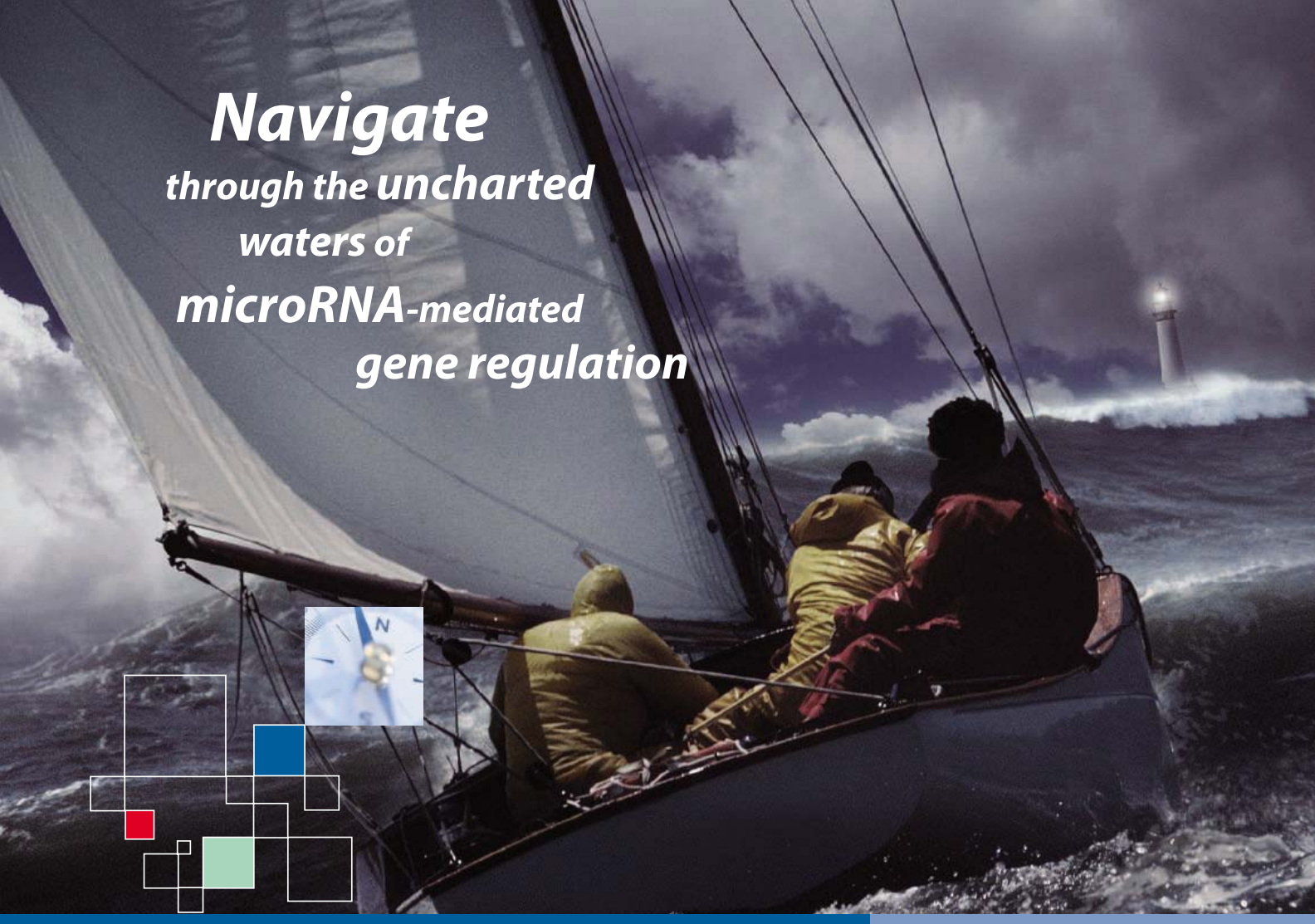

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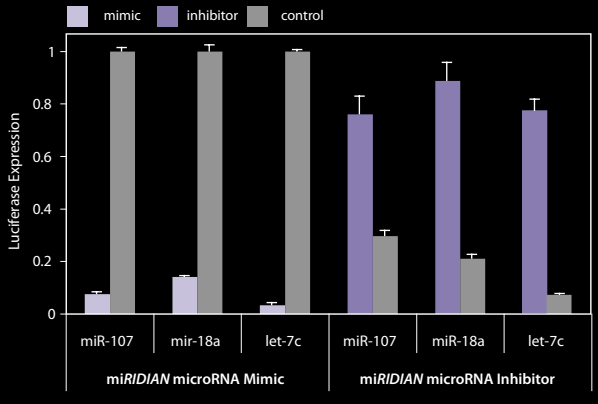

miRIDIAN microRNA Mimic function for three human miRNAs was assayed in HeLa (miR-107 and miR-18a) and HepG2 (let-7c) cells. miRIDIAN microRNA Inhibitor function for three human miRNAs was assayed in HeLa (let-7c) and HEK293 (miR-107 and miR-18a) cells. Control samples received no mimic or inhibitor.

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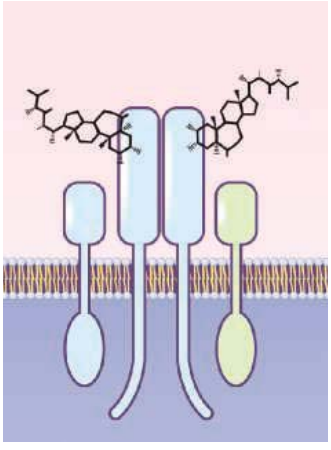
Mother finches lay female eggs before male eggs to safeguard against bloodsucking mites.

Laser on a Chip
New silicon laser could dramatically boost computing speeds.

Hot Flies, Good Times
Study helps explain why alcohol tolerance decreases as temperature rises.



Brassinosteroid signaling starts at the plasma membrane.

SCIENCE'S **STKE**

www.stke.org SIGNAL TRANSDUCTION KNOWLEDGE ENVIRONMENT

PERSPECTIVE: Tumor Suppression by p53 Is Mediated in Part by the Antiangiogenic Activity of Endostatin and Tumstatin
J. Folkman
p53 inhibits not only tumor cell proliferation and survival but also tumor angiogenesis.

PERSPECTIVE: Advances in Understanding Brassinosteroid Signaling
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Plants use plasma membrane receptor complexes to trigger the response to steroid hormones.

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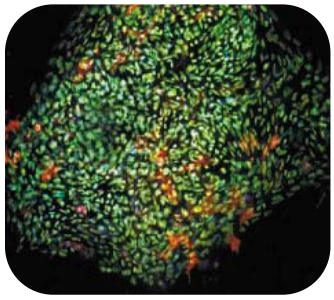

Stem cells and career pressure.

SCIENCE **CAREERS**

www.sciencecareers.org CAREER RESOURCES FOR SCIENTISTS

US: Embryonic Stem Cell Research—Can Young Researchers Succeed?
S. A. Webb
Political uncertainty and funding pressures shape the climate for embryonic stem cell researchers.

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I. S. Levine
Learn how to minimize the personal and professional costs of bullying bosses.

US: Educated Woman, Chapter 55—What a Long, Strange Trip It's Been
M. P. DeWhyse
Micella reflects on the good, the bad, and the ugly of her graduate-school experience.

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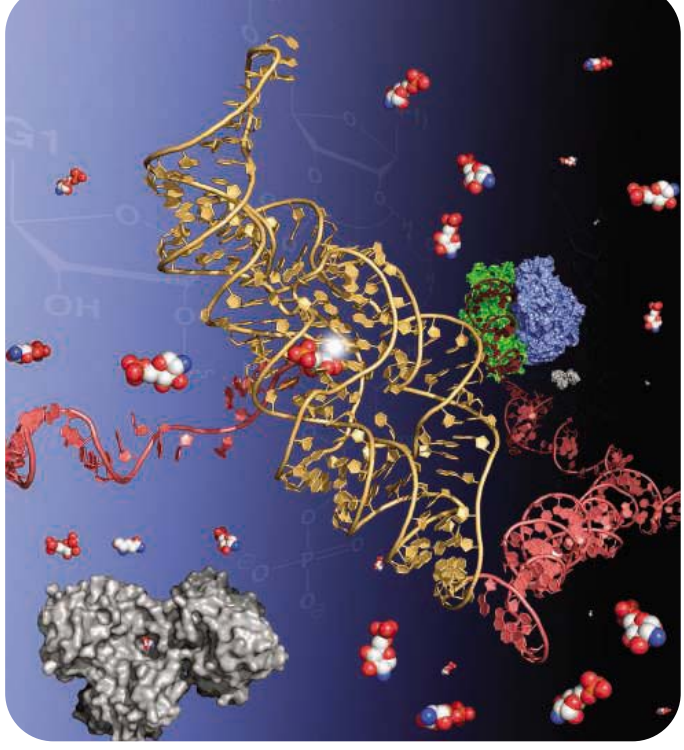
EDITED BY STELLA HURTLEY AND PHIL SZUROMI

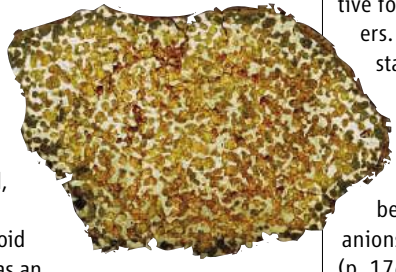


Ribozymes Lost and Found

It has been suggested that self-cleaving RNAs and other ribozymes represented a step—the RNA world—in the origin of life (see the Perspective by **Been**). Now **Klein and Ferré-D'Amaré** (p. 1752) report crystal structures of the *glmS* ribozyme, which regulates the synthesis of glucosamine-6-phosphate (GlcN6P), a key metabolic precursor of the bacterial cell wall. The structures cover the precleavage state, both unbound and bound to the competitive inhibitor glucose-6-phosphate, and the postcleavage state. Unlike other riboswitches, where metabolite binding regulates activity by inducing a conformational change, in GlmS the ribozyme conformation is similar in all three states. GlcN6P binds to a preformed site and is precisely positioned to serve as a coenzyme. Few self-cleaving ribozymes have been detected in mammals, leading to speculation that they have been lost over evolution. **Salehi-Ashtiani *et al.*** (p. 1788) identified a self-cleaving ribozyme in the human genome that shares biochemical and structural properties with hepatitis delta virus ribozymes.

Spawned from Vesta

Meteorites offer glimpses of the earliest stages of planetary formation. Stony-iron meteorites come in two main classes, pallasites and mesosiderites, and it was thought they may have had similar origins. **Greenwood *et al.*** (p. 1763, published online 24 August 2006; see the Perspective by **Clayton**) have found that their oxygen isotope properties differ, suggesting they come from distinct places. The characteristics of mesosiderites suggest they came from the third largest asteroid, Vesta, the target of the NASA Dawn Mission. Pallasites are made of mixed core-mantle material from a disrupted asteroid, indicating that extensive asteroid deformation was an integral part of planetary accretion in the early Solar System.



All in One Shot

Ultrafast laser studies have relied on one light pulse to initiate a chemical reaction and a second one to probe the outcome. Dynamics are measured by continuously repeating this process while successively lengthening the time between the two pulses. Because this approach requires many laser shots, easily depleted samples such as ordered crystals often decompose before sufficient data can be acquired. **Poulin and Nelson** (p. 1756, published online 31 August; see the Perspective by **Apkarian**) developed a scheme in which a delay gradient across a single femtosecond probe pulse allows chemical events spanning a 10-picosecond period to be tracked with subpicosecond resolution. They measured the photodissociation and recombination dynamics of crystalline I_3^- and find that geminate recombination rates depend sensitively on the crystal structure.

A Small Advantage

Capacitors work by storing charge in conductive foils separated by dry nonconducting layers. Supercapacitors also store charge in a static form, but resemble batteries in that they use porous conductors and an electrolyte to store and conduct the charges. It has been assumed that larger pores should lead to better performance because they increase the mobility of the anions and cations. However, **Chmiola *et al.*** (p. 1760) now show an anomalous increase in capacitance for pores smaller than 1 nanometer that may allow development of supercapacitors with higher energy densities.

Earthquakes Unzipped

Earthquake rupture has long been thought to occur by propagation of a crack, but more recent observations and theory seem to indicate a "pulse-like" or "self-healing" mode of rupture propagation. In a series of model experiments, **Lykotrafitis *et al.*** (p. 1765; see the Perspective by **Marone and Richardson**) use a combination of dynamic photoelasticity and laser interferometry techniques to watch various rupture modes propagating along frictionally held, incoherent, interfaces and address the question of what controls slip at a point on a fault during an earthquake in realistic settings. The results show that self-healing pulses are typical and that crack-like or pulse-like modes, or both, can pertain depending on conditions.

All Mixed Down

Turbulence near the surface of the ocean helps transport nutrients to deeper regions and exchange gases with the atmosphere. Most assessments of turbulent mixing have focused on physical drivers, such as wind. **Kunze *et al.*** (p. 1768; see the news story by **Kerr**) report that the dusk ascent of abundant krill (a type of pelagic crustacean) from their daytime depth of 100 m to the surface generates significant turbulence, up to four orders of magnitude greater than that observed at other times, in a coastal inlet. If the effect is widespread, surface mixing could have a significant biological origin.

Icing Up

Cirrus clouds reflect shortwave radiation from the Sun and absorb reflected longwave radiation. The magnitude of these effects depends on the properties of their constituent ice particles and how they form and grow. **Abbatt *et al.*** (p. 1770, published online 31 August) describe that ice can form via heterogeneous nucleation on solid ammonium sulfate aerosols. Prevailing theories have assumed that ammonium sulfate aerosol nucleate ice from the liquid state through a homogeneous process. These findings raise the question of how anthropogenic ammonia emissions, which now exceed natural ones, might impact the formation of upper tropospheric ice clouds.

CREDITS (TOP TO BOTTOM): KLEIN AND FERRÉ-D'AMARÉ; NATURAL HISTORY MUSEUM

Climate and Genetic Change

Some organisms undergo genetic change when they are exposed to higher than normal temperatures. However, whether recent global warming might already be driving such changes has been uncertain. **Balanyá *et al.*** (p. 1773) compiled data on chromosomal polymorphisms covering periods of 13 to 46 years for 26 populations of the fruit fly *Drosophila subobscura* on three continents. Weather records for the same periods and locations suggest that recent climate warming is associated with genetic change in 22 of the populations, favoring genotypes characteristic of low latitudes.

Social Experience and the Need to Sleep

Sleep is widely observed in the animal kingdom and yet we still don't know why it is beneficial. Studying *Drosophila*, **Ganguly-Fitzgerald *et al.*** (p. 1775) developed a strategy for elucidating the mechanisms underlying the need to sleep. They observed that a rich social experience, versus an impoverished one, increased the duration of sleep, which in turn was promoted by processes that underlie learning and memory, such as dopamine and cyclic adenosine monophosphate signaling pathways. Mutations in 17 genes were found to disrupt experience-dependent changes in sleep.

Infectious Amyloid

β-amyloid plays a key role in Alzheimer's disease. There also exist marked pathological similarities between Alzheimer's disease and so-called prion diseases like Mad cow disease. **Meyer-Luehmann *et al.*** (p. 1781) now show that cerebral β-amyloid–amyloidosis can be induced by the injection of exogenous, β-amyloid–rich brain extract, and that cerebral amyloid induction is dependent on intrinsic properties of the injected β-amyloid agent and the host that receives the injection. The results suggest the occurrence of polymorphic β-amyloid species with varying biological activities, reminiscent of prion strains. The findings underscore the commonalities among diseases of protein aggregation and assembly.

CREDIT: MEYER-LUEHMANN *ET AL.*

Tumors Send for Help

Solid tumors require a blood supply for their growth, and they recruit surrounding host endothelial cells to build new blood vessels. The extent to which tumors enlist the help of the endothelial progenitor cells (EPCs) that circulate in the blood has been controversial. Studying mouse models, **Shaked *et al.*** (p. 1785) show that treatment of tumors with drugs called vascular disrupting agents (VDAs) leads to a sudden and dramatic mobilization of EPCs to the tumor rim. When EPC mobilization was prevented, the tumors were more responsive to the therapy. Thus, under certain circumstances, the contribution of EPCs to tumor angiogenesis is indeed substantial.

Protein Pathways in Epilepsy

One cause of epilepsy is mutations in proteins that function in the brain. **Fukata *et al.*** (p. 1792; see the Perspective by **Snyder**) identified the partners of a complex of proteins located at rat brain synapses. Of the various components, one (LGI1) seems to function as a ligand, one (ADAM22) as a receptor, and one (PSD-95) as a scaffolding anchor. LGI1 controls the strength of excitatory synapses. Both the ligand and the receptor of this complex are implicated by genetics and mutations as being causative for certain types of epilepsy.

Beyond Self–Non-Self for MHC

Proteins of the major histocompatibility complex class 1 (MHC1), which are important in identifying self and non-self tissue for the immune system, are also found in the brain. **Syken *et al.*** (p. 1795) show that a receptor, PirB, to which the MHC1 proteins bind, is also found in neurons of the brain. In mice carrying a mutant PirB lacking its signal transduction capabilities, the overall structure of the brain remained normal. However, these mice showed greater than normal plasticity in the visual cortex. Thus, intercellular signaling through PirB seems to be critical for keeping visual plasticity within limits.

Q AAAs

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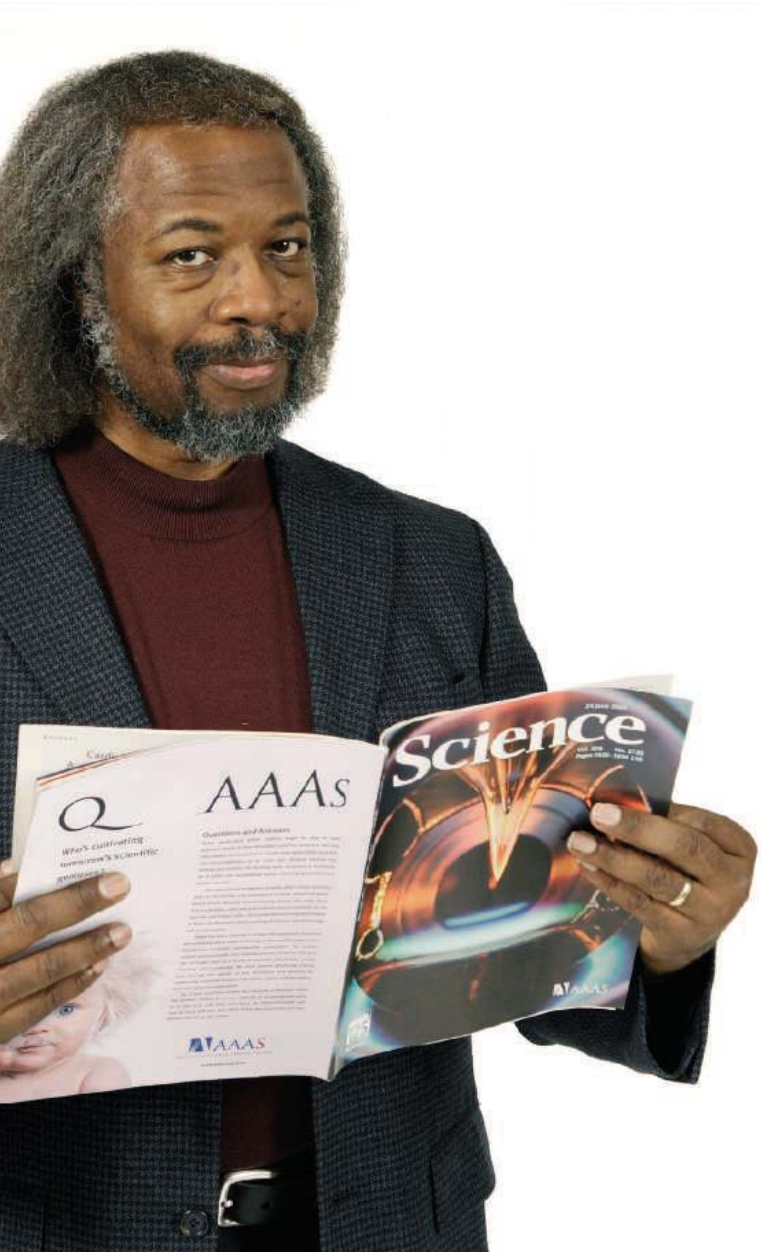

"As a child I got very interested in space travel. When I was six my father gave me some books on rockets and stars. And my universe suddenly exploded in size because I realized those lights in the sky I was looking at were actually places.

I wanted to go there. And I discovered that science and technology was a gift that made this possible. The thrill of most Christmas presents can quickly wear off. But I've found that physics is a gift that is ALWAYS exciting.

I've been a member of AAAS for a number of years. I think it's important to join because AAAS represents scientists in government, to the corporate sector, and to the public. This is very vital because so much of today's science is not widely understood.

I also appreciate getting *Science* because of the breadth of topics it covers. It gives me a great grounding for many activities in my professional life, such as advising government agencies and private corporations."

Jim Gates is a theoretical physicist and professor at the University of Maryland. He's also a member of AAAS.

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S. James Gates Jr., Ph.D.
Theoretical physicist
and AAAS member

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Elizabeth G. Nabel, M.D. is director of the National Heart, Lung, and Blood Institute at NIH in Bethesda, MD. Her scientific research concerns the molecular genetics of cardiovascular disease. E-mail: nabele@nhlbi.nih.gov

The Women's Health Initiative

EARLIER THIS YEAR, AFTER 12 YEARS, 7.5 MILLION FORMS, AND 1 MILLION CLINIC VISITS, the Women's Health Initiative (WHI), a major 15-year research program of the U.S. National Institutes of Health (NIH), announced its research findings about women and chronic diseases. It indicated that certain interventions to treat cardiovascular disease, cancer, and osteoporosis were not as beneficial as thought. Conventional wisdom appeared to have been stood on its head, provoking strong reactions among scientists and the public: disbelief, disagreement, discouragement, and a fair measure of dissention and disharmony. Upon reflection, the results are reasonable, but we learned some lessons about how to clarify the broad application of findings as complex as those of the WHI. Now, in preparing to further delve into this rich resource of participant data, the WHI can make the most of an unprecedented opportunity to understand the mechanisms by which disorders in women develop, how they can be prevented, and how interventions can confer benefits or risks.

Launched in 1991, the WHI reflected increasing attention to women's health and a strong demand for reliable information to guide their health care decisions. It is the first broad-scale examination of the major causes of disability and death among postmenopausal women, recruiting more than 161,000 volunteers in the United States between 50 and 79 years of age. Clinical trials tested three interventions: hormone therapy to prevent coronary heart disease and osteoporotic fractures, a reduced-fat diet to prevent breast and colorectal cancers and coronary heart disease, and calcium and vitamin D supplementation to prevent fractures and colorectal cancer.

The hormone trials were prematurely halted when an unfavorable risk/benefit profile indicated that estrogen-based therapies increased the risk of coronary heart disease, stroke, and breast cancer. The other trials failed to definitively establish the merits of their interventions. On the positive side, certain subgroups derived a benefit regarding breast cancer and bone health. The conclusion was that there may be a role for low-fat diets or calcium and vitamin D supplementation in preventing some chronic diseases.

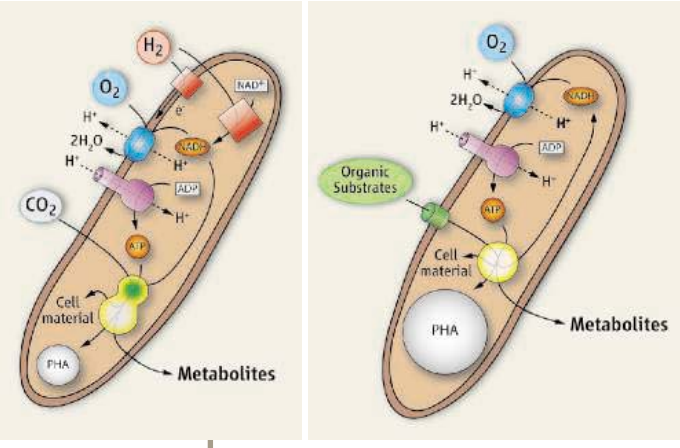
Should we be surprised by the WHI results? I think not. The study identified strategies that had been correlated with beneficial outcomes among selected cohorts of women and then tested the efficacy of these strategies in a huge group of volunteers representing a range of ages, backgrounds, and experiences. This was all quite reasonable and entirely concordant with NIH's public health mission, but it was probably naïve to expect results that would be broadly applicable to such a diverse group.

On the contrary, it makes sense to expect that the interventions may be beneficial (or harmful) only among woman with particular genetic, biological, and/or environmental characteristics. It is precisely this issue that will be the focus of the next chapter of the WHI. We have solicited proposals to mine the WHI data to identify genes and biological markers that might explain the pathways of disease development as well as the effects of treatment on disease outcomes. For example, genetic polymorphisms in a particular blood coagulant (factor V Leiden) increase the risk for venous thrombosis; hormone therapy also increases the thrombotic risk in some women. We are eager to understand the level of thrombotic risk for women with a genetic susceptibility to thrombosis when exposed to environmental and treatment factors, such as hormone therapy. These research findings would have direct implications for treatment options.

It is important that the first chapter of the WHI study emphasized examining the biological differences between women and men. But I believe there is equal or even greater merit in examining individual biological variability—how women differ from one another—to understand why a given woman may fall ill and how we can best make her well. This knowledge is an essential prerequisite to the development of prevention and treatments that are tailored to the unique personal characteristics and health needs of each woman. Our investment in the WHI will yield untold rewards to women worldwide if we succeed, and this is exciting news for all women.

– Elizabeth G. Nabel

10.1126/science.1134995



Metabolic schematic of *Ralstonia*.

BIOTECHNOLOGY

A Plastic Genome

Ralstonia eutropha H16 is a bacterium that can adjust to life on a variety of nutrients (as carbon and energy sources) and can survive periods of anoxia. Two skills are of particular interest: the ability to perform the Knallgas reaction and the storage of carbon in polyhydroxyalkanoate (PHA) granules. The former refers to the explosive combination of H_2 and O_2 (in a 2:1 ratio), which *Ralstonia* carries out in a traditional respiratory fashion, passing protons and electrons separately through membrane-bound carriers until they are added to O_2 in a terminal oxidase complex to produce water. The latter was first detailed almost half a century ago and has led to the biodegradable thermoplastic Biopol and to polythioesters.

Pohlmann *et al.* report the sequence of the two *Ralstonia* chromosomes (~7 Mb), providing an inventory of the many candidate enzymes involved in the synthesis, polymerization, depolymerization, and catabolism of PHAs. The large number of genes encoding β-ketothiolases and acetoacetyl-CoA reductases offers the potential for tinkering with substrate specificity to create an intracellular library of three- to five-carbon hydroxyacid monomers. — GJC

Nat. Biotechnol. **24**, 10.1038/nbt1244 (2006).

BIOMEDICINE

A Pick-Me-Up for Cancer

A major avenue that is being explored in the treatment of cancer is the possibility of mobilizing the immune system to attack tumor cells. However, for reasons that are only slowly becoming clear, encouraging immune cells to destroy tumors remains relatively inefficient.

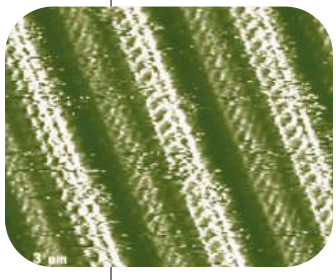
Ohta *et al.* provide evidence that tumors protect themselves from immune attack via extracellular adenosine generated within the hypoxic environment of the tumor mass itself. Previous studies have suggested that during inflammation, the activation of the adenosine receptor (A2AR) on T cells leads to levels of intracellular cyclic AMP that inhibit cell function. In the current experiments, 60% of mice lacking A2AR rejected their tumors, as compared to unimpaired tumor growth seen in mice with immune cells able to signal through the receptor. A2AR antagonists—including caffeine—had similar, albeit less robust, tumor-inhibiting effects that depended on interferon-γ–producing $CD8^+$ T cells. These results lend support to the contested notion that the immune system continuously monitors for malignancy and raises the question of an A2AR-mediated contribution to early spontaneous tumor growth. If this is the case, then inhibition of this pathway might be helpful as an adjunct to immune-based therapies for some cancers. — SJS

Proc. Natl. Acad. Sci. U.S.A. **103**, 13132 (2006).

CHEMISTRY

Stand Up, Line Up, Charge!

The miniaturization of electronic devices to the nanometer scale requires the fabrication of extremely narrow wires. One approach has focused on the synthesis of conducting metal or carbon nanotubes. A second approach is the self-assembly of small molecular components into conduits held together by noncovalent interactions. Stacked aromatic molecules such as tetrathiafulvalene (TTF) could potentially achieve this function. However, when adsorbed on graphite, the highly conjugated TTF molecule interacts strongly with the substrate and lies flat, which minimizes interactions between molecules; hence, the stack motif is unstable. Puigmartí-Luis *et al.* have derivatized TTF by capping two of the terminal sulfur atoms with amide groups, which are in turn bonded to long alkyl chains. The intermolecular hydrogen-bonding interactions between the amides allow the TTF moieties to form long one-dimensional chains in which the π electron–rich cores are tilted at a high angle to the surface. Scanning tunneling microscopy revealed that parallel wires are formed, spaced ~5 nm apart, which is consistent with distances predicted by molecular modeling. Both quantum mechanical calculations and scanning tunneling spectroscopy suggest that the nanowires should be highly conducting. Furthermore, rectifying behavior was observed in the –1- to 1-V range, with a 10-fold increase in current at negative versus positive substrate bias. — PDS



TTF stacks.

J. Am. Chem. Soc. **128**, 10.1021/ja0640288 (2006).

MATERIALS SCIENCE

Negative Index Made Easy

The realization of designer materials, or metamaterials, in which the electrical permittivity and magnetic permeability can be made negative simultaneously has generated much interest, primarily due to theoretical proposals for remarkable applications such as perfect lenses and, most recently, the ability to hide, or cloak, objects from electromagnetic radiation. After the initial demonstration in the microwave regime, much of the experimental effort in metamaterial design has focused on pushing the response of these materials toward shorter wavelengths. However, the design of choice, a split ring resonator coupled to a metal wire, is somewhat limited when it comes to size reduction, and other approaches are being pursued. Chettiar *et al.*

CREDITS (TOP TO BOTTOM): POHLMANN *ET AL.*, *NAT. BIOTECHNOL.* **24**, 10.1038/NBT1244 (2006); PUIGMARTÍ-LUIS *ET AL.*, *J. AM. CHEM. SOC.* **128**, 10.1021/JA0640288 (2006)

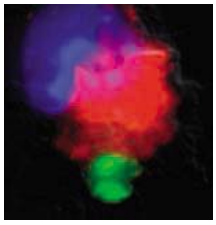
propose the use of a simpler structure to achieve negative refraction: a pair of metallic nanostrips separated by a thin dielectric layer, easily fabricated with existing deposition and lithographic technology down to feature sizes in the 10- to 100-nm range. Their simulations show that coupling such pairs of nanostrips to continuous metal films should provide a negative refractive index in the optical and infrared regimes. — ISO

Opt. Express **14**, 7872 (2006).

CELL BIOLOGY

Stripped and Eliminated

Protozoan parasites such as *Plasmodium* and *Toxoplasma* invade host cells and divide within a parasitophorous vacuole. The vacuolar membrane is modified by the invading parasite in order to forestall its fusion with host endocytic and degradative organelles (lysosomes). Ling *et al*. have examined how mouse macrophages, after being infected by *T. gondii*, can break through this parasite-constructed defensive wall. Cells from mice lacking an interferon-γ–inducible p47 GTPase (IGTP) failed to eliminate the pathogen. In contrast, in wild-type cells the parasitophorous vacuole membrane was disrupted during the degradation process, and the parasite plasma membrane was stripped away. The parasite was then engulfed by a double-membrane autophagosome, which fused with lysosomes, leading to destruction of the parasite. Recently, IGTPs have been shown to play a similar role in the elimination of intracellular *Mycobacterium* in mice and in humans (Singh *et al*., Reports, 8 September 2006, p. 1438). — SMH



Lysosomes (red) do not fuse with invasive *Toxoplasma* (green) in cells lacking IGTP.

CREDIT: YUN LING AND GEORGE YAP

J. Exp Med. **203**, 2063 (2006).

ASTROPHYSICS

Signs of Collapse

When their nuclear fuel is exhausted, stars die, and the residual iron core collapses on itself. The outcome of a star's death throes depends on mass, however. Stars with between 10 and 20 times the mass of the Sun collapse in a spectacular explosion known as a supernova, leaving behind a neutron star, whereas those larger than 20 solar masses implode to form black holes in a "hypernova." In both cases, copious bursts of neutrinos are released along with optical, x-ray, and gamma radiation. Most scenarios for hypernova collapse involve rapidly rotating stars, but recent studies indicate that some massive stars may be rotating only slowly or not at all.

Sumiyoshi *et al.* have carried out simulations showing that such stars may lead to explosions that are very dim in the electromagnetic spectrum, but that still lead to black hole formation and powerful neutrino bursts. These neutrino signals are sensitive indicators of the equation of state of matter in the collapsed star. (The equation of state relates basic quantities such as pressure, density, and temperature.) As a result, these neutrino bursts could offer a valuable diagnostic tool for studying the properties of stellar matter. — DV

Phys. Rev. Lett. **97**, 091101 (2006).

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<< An Open and Shut Case

Stomata are openings on the surfaces of leaves that mediate the exchange of gases, which is essential for respiration and osmotic balance. However, these doorways also provide a route by which infectious bacteria can gain access to plant internal tissues. Stomata open and close in response to changes in exposure to light, humidity, and other stimuli, but Melotto *et al.* show that they can also be shut as a defense against bacterial invasion. *Arabidopsis* closed their stomata within 2 hours of exposure to the pathogenic bacterium *Pseudomonas syringae*, but reopened them a few hours later. Microscopic observation showed that the bacteria were able to detect and migrate toward open stomata, perhaps sensing nutrients or other molecules released from the plant interior. Flg22, a peptide derived from the bacterial flagellin protein, or lipopolysaccharide, a component of the bacterial outer cell wall, could trigger stomatal closure, and plants are known to have immune receptors that recognize these molecules. The subsequent reopening of the stomata led the authors to test whether *P. syringae* produced a virulence factor that could override the host plant's protective mechanism. Indeed, they found that the bacterially produced polyketide toxin coronatine was required for reopening of the stomata. These results reveal that plants have developed an innate immune mechanism to protect themselves against bacterial invasion and that in response some bacteria have developed a virulence factor that reopens doors. — LBR

Cell **126**, 969 (2006).

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Information for Contributors

See pages 102 and 103 of the 6 January 2006 issue or access www.sciencemag.org/feature/contribinfo/home.shtml

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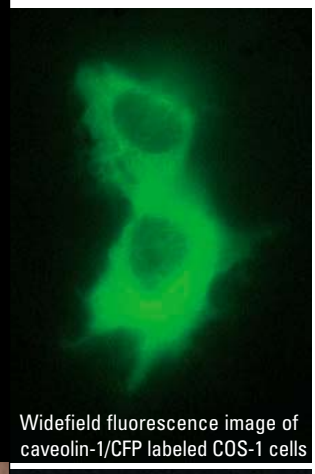
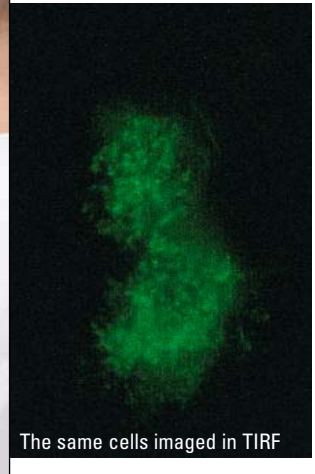


Widefield fluorescence image of caveolin-1/CFP labeled COS-1 cells



The same cells imaged in TIRF

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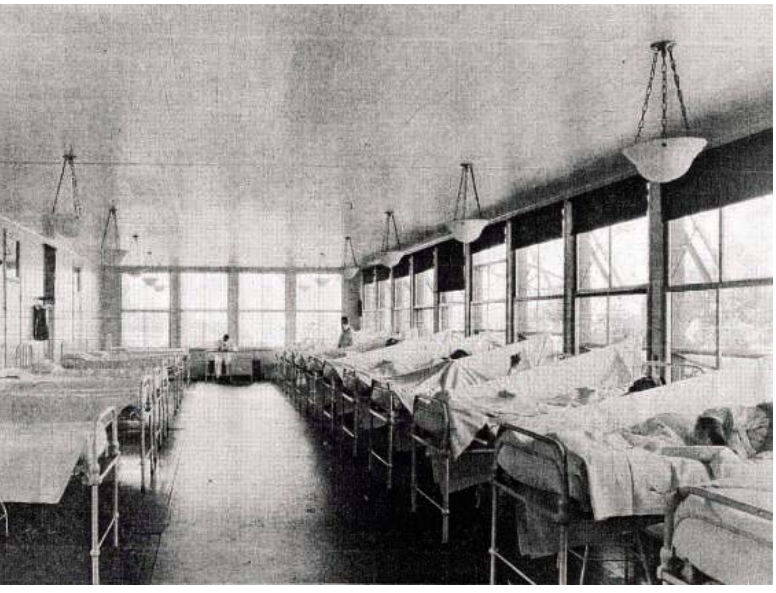


DOCUMENTS

<< Pandemic Lessons Learned

Princeton University in New Jersey forbade students from leaving campus and ringed the dorms with sentries. Gunnison County in Colorado closed its schools for more than 3 months, banned public gatherings, and quarantined visitors. Measures like these might seem extreme, but they apparently kept influenza at bay during the 1918–1920 pandemic. This new archive from the University of Michigan Medical School in Ann Arbor details the responses of seven such "escape communities" that suffered no more than one flu death. The site is based on a recent report commissioned by the U.S. Defense Threat Reduction Agency to help prepare for future pandemics. It includes contemporary newspaper accounts, letters, and other documents that reveal the tenor of the times. Left, the pneumonia ward at the San Francisco Naval Training Station on Yerba Buena Island, which was one of the escape communities. >>
www.med.umich.edu/medschool/chm/influenza/index.htm

ARCHIVE

The Royal Treatment

Since it began publishing in 1665, Britain's Royal Society has run works by Newton, Robert Hooke, Michael Faraday, Watson and Crick, and plenty of other scientific giants. For the next 2 months, visitors can troll the society's complete journal archive and download articles for free. Historically important publications stowed here include astronomer Edmund Halley's account of the eponymous comet and a description of Benjamin Franklin's kite-flying experiment. Free access ends in December. >> www.pubs.royalsoc.ac.uk/archive

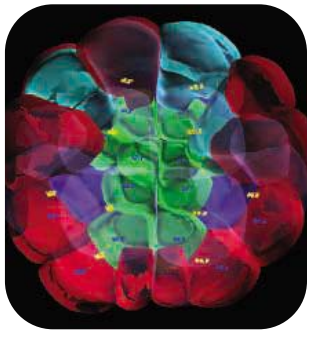
EXHIBITS

Not Just a Guy Thing

At Changing the Face of Medicine, meet some of the doctors who shattered the stereotype of the M.D. as a middle-aged man with a stethoscope and a Thursday tee time. The National Library of Medicine exhibit tells the life stories of more than 200 women physicians in the United States. You can cue up video interviews with living doctors and peruse biographies of historical figures such as Elizabeth Blackwell (1821–1910), the first woman M.D. in the country. Visitors can also try out interactive features such as a pioneering design by Mary Putnam Jacobi (1842–1906) for a sphygmograph, a device that measures pulse strength. >> www.nlm.nih.gov/changingthefaceofmedicine

DATABASE

Path to a Tunicate



The filter-feeding marine animals called ascidians, or tunicates, have sucked in evolutionary and developmental biologists. The fascination stems in part from the creatures' close kinship to vertebrates and their simple embryos, which serve as good models for development. Hosted by French and Japanese labs, ANISEED* is packed with embryological and molecular information on ascidians. With free visualization software, you can pick an embryo such as the 44-cell stage of *Ciona intestinalis* (right) and highlight developmental lineages or pinpoint areas of contact between cells. The site also houses gene-expression data from in situ hybridization experiments. To enjoy some pretty photos, visit the Dutch Ascidians Homepage.† Graduate student Arjan Gittenberger of the National Museum of Natural History in Leiden, the Netherlands, has corralled shots of more than 100 ascidian species from around the world. >>
* crfb.univ-mrs.fr/aniseed/index.php † www.ascidians.com

RESOURCES

Life on the Subcontinent

Indian villagers brew a tonic from the bark of the neem tree (*Azadirachta indica*; above), dine on its shoots, and use its twigs for toothbrushes. The multipurpose tree is also one of the species listed in the Indian Bioresources Information Network, hosted by the country's Department of Biotechnology. The growing site inventories India's plants, animals, and microbes. Visitors can browse the fish catalog by scientific name and by common name in 20 languages. The more than 1800 species accounts offer taxonomic summaries, descriptions, range maps, and images. Another collection furnishes similar data on more than 3000 medically or economically significant plant species. Sections on land vertebrates and other groups are under construction and can be balky. >> www.ibin.co.in

Send site suggestions to >> netwatch@aaas.org **Archive:** www.sciencemag.org/netwatch

CREDITS (TOP TO BOTTOM): UNIVERSITY OF MICHIGAN MEDICAL SCHOOL; WILLIAM M. CIESLA/FOREST HEALTH MANAGEMENT INTERNATIONAL; PATRICK LEMAIRE AND OLIVIER TASSY

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MORE NEW SPECIES IN NEW GUINEA

This epaulette shark (*Hemiscyllium freycineti*), which pulls itself along the ocean floor on its fins, is one of about 50 new species spotted during a survey led by Washington, D.C.–based Conservation International in a rich coral reef area off the coast of Papua, the western (Indonesian) half of New Guinea. Findings were announced by lead researcher Mark Erdmann on 18 September in Jakarta.

Stem Cell Agreement

The Wisconsin group that owns 13 of the 21 human embryonic stem (hES) cell lines used by federally funded researchers says it will start supplying new lines that purport to offer an alternative to embryo destruction—*if* the government finds them eligible for funding.

The lines were derived by Advanced Cell Technology (ACT) in Alameda, California, using a process described last month in a paper in *Nature*: taking single cells, or blastomeres, from early embryos, which in theory could be done without harming the embryos (*Science*, 25 August, p. 1031). ACT would scale up production of the cells, and WiCell Research Institute in Wisconsin would test and distribute them.

National Institutes of Health stem cell czar James Battey says that for ACT's cells to pass muster, it would have to be shown that blastomeres cannot develop into embryos, because federal researchers are not allowed to experiment with human embryos. And, he adds, "no one can say with complete certainty that there is no risk to the embryo that remains." Also, NIH would have to get a legal opinion on whether such cells are kosher under President George W. Bush's directive that no work with hES cell lines derived after 9 August 2001 can be federally funded.

Scientists agree that even if ACT's process works, it would be no substitute for legislation allowing federally funded researchers to use new lines derived from leftover embryos created at fertility clinics.

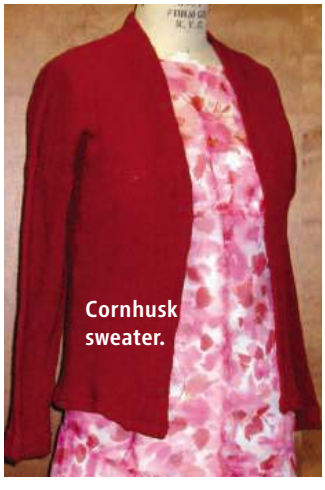
Sex and Death

The motivations of suicide bombers differ depending on their sex, says a researcher at the University of Virginia, Charlottesville. Psychiatrist J. Anderson Thomson Jr. says that whereas males see themselves as part of a larger entity, females seem more propelled by individual motives.

Male suicide attackers are not lone losers but members of tightly knit bands bound by ties of rage and religion. Their behavior is consistent with our ancient history of "male-bonded coalitionary violence," involving "lethal raids" practiced by small bands against their enemies, argues Thomson.

But women do not fit this pattern. In a paper delivered at the biennial meeting of the International Society for Human Ethology in Detroit, Michigan, last month, Thomson mentioned Chechen, Palestinian, and Hindu female suicide terrorists who had been shunned for adultery or because they had been raped, divorced because of infertility, or whose husbands or brothers had been murdered by the enemy. In these cases, he asserts, the motives have more to do with shame or personal revenge than a larger cause. And rather than being motivated by bonds with their fellows, Thompson added, all these women were "recruited, trained, directed, or in some manner controlled by men."

Brian Jenkins, a longtime terrorism expert at the RAND Corp. in Santa Monica, California, says that although the paper offers only anecdotal evidence, it contains "some interesting insights. … There clearly is a sex difference."



Cornhusk sweater.

CREDITS (TOP TO BOTTOM): GERRY ALLEN; HANDOUT/REUTERS/CORBIS; Y. YANG

<< AGRI-COUTURE

More than half of the 67 million tons of textile fibers produced annually are petroleum-based synthetics. But with rocketing oil prices, agricultural byproducts are gaining attention as natural fiber sources, scientists reported last week at the American Chemical Society meeting in San Francisco, California.

Textile scientist Yiqi Yang of the University of Nebraska, Lincoln, said he has gotten fibers from rice straw that are "long and fine enough for textiles but still very strong." Using alkali and enzymes, he and student Narendra Reddy extracted finger-length fibers that they say rival linen and cotton in flexibility and strength. Adding cotton, they spun a yarn and wove it into rice/cotton fabric. Yang estimates that 58 million tons of textile fiber could be produced from half of the 580 million tons of waste rice straw grown each year. Brian George, a textile engineer at Philadelphia University in Pennsylvania, says the relative stiffness of such fibers makes them hard to work with unless they are blended with cotton or flax, but that the idea seems economically viable if the fibers "can be processed on standard textile equipment."

Yang says rice-straw fibers are stronger than those from cornhusks, which he managed to make a sweater out of a few years ago. His next project is to get spinnable fibers from chicken feathers, whose honeycomb structure, he says, could potentially make for textiles lighter and warmer than wool.

GM rice mystery
1714

Baby "Lucy"
1716

GENDER ISSUES

Universities Urged to Improve Hiring and Advancement of Women

U.S. universities foster "a culture that fundamentally discriminates against women," says a new report by the National Academies on the status of women in academic science and engineering. Their underrepresentation is "deeply troubling and embarrassing," according to the report, which suggests that institutions create a body to collect data, set standards, and ultimately monitor compliance to increase the number of women in technical fields.

Beyond Bias and Barriers: Fulfilling the Potential of Women in Academic Science and Engineering cites research demonstrating that women are paid less, promoted more slowly, bypassed for honors, and subjected to implicit gender bias from both their male and female colleagues. The 18-member panel—chaired by Donna Shalala, president of the University of Miami in Coral Gables, Florida, and made up primarily of female university presidents, provosts, and senior professors—also finds no scientific basis to the argument that inherent differences between the genders are at the root of the problem. "This report confronts the myths; it is a data and information-driven study," says Donna Dean, a biochemist and former National Institutes of Health official who is senior science adviser with the Washington lobbying firm Lewis-Burke Associates. But others, such as chemist and activist Debra Rolison of the Naval Research Laboratory in Washington, D.C., expressed disappointment that the panel didn't come out more strongly for aggressive use of existing federal laws.

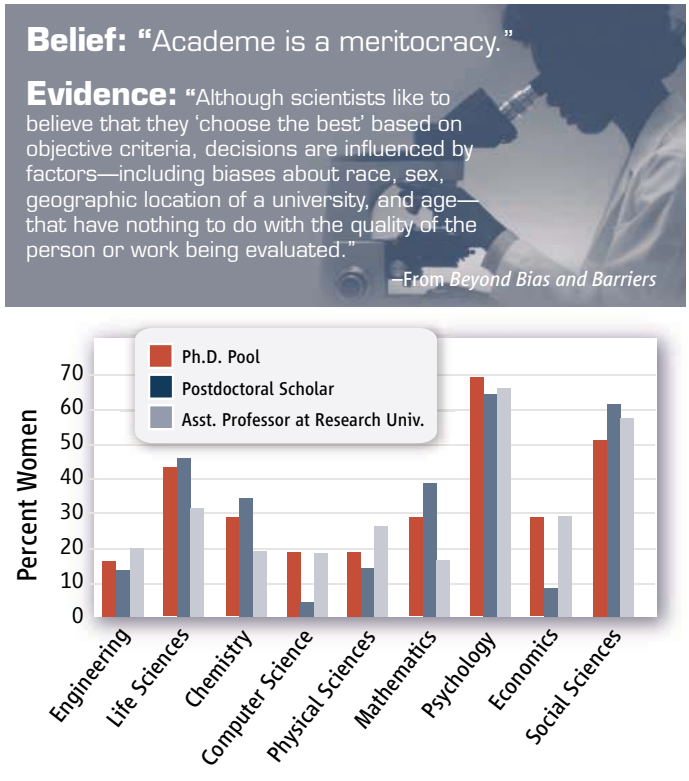
The 18 September report is the latest in a series of private and government studies examining the status of women in senior science and engineering positions across the country. One of its few concrete proposals is an "interinstitution monitoring organization" to set norms for expanding the role of women in the sciences and engineering. The organization, the panel suggests, would be similar to the National Collegiate Athletic Association, which serves as an intermediary between universities and federal agencies. The American Council on Education has agreed to convene several national education organizations to "define the scope and structure of data collection," says ACE Vice President Claire Van Ummersen. "This would be a way for the profession to police itself," says Nancy Hopkins, a biologist at the Massachusetts Institute of Technology (MIT) in Cambridge, who chaired a study in 1999 focusing on the problem at her university.

But Rolison criticized the panel for not demanding greater accountability. That includes strict enforcement of a 1972 law, popularly known as Title IX, that prohibits any education program or activity receiving federal funding from denying equal benefits to women. "That's the missing piece," she says. Shalala acknowledged at a press conference that the federal government has spent more time and energy ensuring equity on collegiate playing fields than in the laboratory. "There are laws on the books which are not being enforced," she added. Shalala later told *Science* that the report's focus is "not any individual law, but *all* enforcement. … Institutional leaders and professional societies have to make systemic changes to provide opportunities."

The fundamental problem, the panel notes, is not attracting women into science but retaining them once they are trained. "The pipeline is in better shape than I thought," says Ana Mari Cauci, a panel member and a psychologist at the University of Washington, Seattle. At MIT, for example, more than half of science undergraduates are female, and more than one-third of engineering students are women. "It is not lack of talent but unintentional biases and our outmoded institutional structures that are hindering the access and advancement of women," the report states. For example, the report says the culture still favors academics with a stay-at-home spouse—typically a wife. Fewer than half the spouses of male faculty members in the sciences are employed full-time, whereas 90% of the husbands of women faculty members work outside the home.

The gap widens with seniority, the report notes. At leading research universities, fewer than 15% of full professors in the life sciences are women, and in the physical sciences, that figure remains in the single digits. "Women from minority racial and ethnic backgrounds are virtually absent from the nation's leading science and engineering departments," the study adds.

The panel dedicated the report to Denice Denton, a panel member and chancellor of the University of California, Santa Cruz, who committed suicide in June (*Science*, 30 June, p. 1857). **–ANDREW LAWLER**



Draining the pool. Women scientists are underrepresented at the entry level within certain disciplines in academia.

SOURCE: NATIONAL SCIENCE FOUNDATION (2006) SURVEY OF DOCTORAL RECIPIENTS (2003)

Bosnia's pyramid frenzy 1718

Chinese part-time profs 1721

Threat to African flamingos 1724

GEOLOGY

Katrina Study Stirs Debate on Coastal Restoration

A maverick ecologist is suggesting that some of the massive and costly engineering fixes being used to restore coastal wetlands in Louisiana will barely make a dent in the problem.

In a paper published online by *Science* this week (www.sciencemag.org/cgi/content/abstract/1129116), Eugene Turner of Louisiana State University in Baton Rouge and three LSU colleagues report the first coastwide study of sedimentation from hurricanes Katrina and Rita. They conclude that hurricanes are by far the most important source of inorganic sediments in the wetlands, dumping so much that, in comparison, costly schemes to channel sediment-bearing Mississippi River water back to the wetlands will have a "trivial" effect. Instead, they argue, restoration efforts should focus on restoring the buildup of organic material.

Many of Turner's counterparts in the ecological community disagree, however, saying that although the researchers have marshaled useful new data, the measurements don't justify their conclusions. "It would be very unwise to use this study to overthrow the thinking about coastal restoration," says physical geographer Torbjörn Törnqvist of Tulane University in New Orleans, Louisiana. "We need to be extremely careful" about interpreting the new results.

Most experts trace Louisiana's coastal degradation in large part back to the levees that were built in the 20th century to control the Mississippi River (*Science*, 25 November 2005, p. 1264). They contend that the levees prevented floods from delivering the necessary silt to delta wetlands. As a remedy, the state of Louisiana and the federal government spent $145 million to construct a pair of prototype structures in 1991 and 2002 to divert river water into wetlands. Several more structures are proposed in a bill before Congress.

But hurricanes also dump mud and debris onto wetlands, and Turner had long suspected that hurricanes might be an even bigger source of sediment than the mighty Mississippi. Katrina and Rita gave him a chance to find out.

Using a rapidly awarded grant from the National Science Foundation, Turner and his colleagues chartered a helicopter in early November 2005 and took samples of storm-surge deposits from 186 sites across 38,588 square kilometers of coastal wetlands. They found plenty of muck. On average, the muddy sediment was 5 centimeters thick; that means Katrina and Rita left a combined 130 million metric tons of sediment on the wetlands, Turner and his colleagues calculate.

Muck galore. Hurricane Katrina covered coastal wetlands with an abundance of silt. Based on new measurements, some researchers argue that hurricanes provide almost all the inorganic sediment the ecosystem needs.

CREDIT: R. E. TURNER

Other ecologists welcome these new measurements, but they take exception to Turner's next, critical, assumptions. Based on a scanty historical record, Turner and his colleagues estimate that storms with a surge as big as Katrina's hit the Louisiana coast on average every 7.9 years. At that frequency, hurricanes deposit about 26 million metric tons of sediment a year on wetlands and associated open water—more than five times the amount contributed by the Mississippi River floods before the levees were constructed, Turner and his colleagues calculate.

Turner says his analysis bolsters his contention that Louisiana's wetlands don't face a shortage of inorganic sediment. The major cause of wetland loss, Turner has long argued, is canals dug for oil and gas drilling that changed the hydrodynamics of the region. This, in turn, he believes, stunted and killed plants and retarded the buildup of organic materials. He favors filling these canals and restoring adjacent marshes.

Many other experts, however, suspect that Turner has overestimated the sedimentation rate of hurricanes. First, they say, surges probably eroded shallow bays and then dumped the silt on the marsh. "It's robbing Peter to pay Paul," says Joseph Kelley of the University of Maine, Orono. In addition, they believe that major storms such as Katrina strike much less often than Turner and his colleagues estimate.

They also point out that diversion projects not only supply sediment but also help reduce salinity and provide nutrients. In fact, Denise Reed of the University of New Orleans and others advocate constructing an even larger diversion of the Mississippi River, to build substantial new land south of New Orleans. "If you don't have diversions as a major part of your restoration efforts, you can't save the coast," says John Day of LSU, who has studied one of the diversions for a decade. **–ERIK STOKSTAD**

GENETICALLY MODIFIED CROPS

Tracing the Transatlantic Spread of GM Rice

Amid product recalls and plummeting prices, scientists are trying to figure out exactly how traces of an experimental variety of genetically modified (GM) rice ended up in commercially available supplies in the United States and Europe. Although the herbicide-resistant strain was never approved or marketed, traces of it have appeared in samples collected on both continents. Agriculture officials stress that the rice poses no health threat, but its spread is a cautionary tale that introduced genes may be harder to contain than some scientists and industry leaders had hoped. The finds "set a really bad example for genes that we do want to keep contained," says plant geneticist Norman Ellstrand of the University of California, Riverside.

The variety, called Liberty Link 601 (LL601), was grown in test plots in several states between 1998 and 2001. Designed to be resistant to the broad-spectrum Liberty herbicide sold by Aventis CropScience (later bought by the German company Bayer), it was not as successful as hoped, and Aventis discontinued research on the strain in 2001. In late July, Bayer notified the U.S. Department of Agriculture (USDA) that it had found traces of LL601 in commercial samples of long-grain rice stored in Arkansas and Missouri. When USDA announced the find two and a half weeks later, U.S. rice prices fell by nearly 10% in 2 days.

On 11 September, European Union officials confirmed that 33 of 162 samples tested by rice millers across Europe, a major importer of U.S.–grown rice, had shown traces of LL601. Officials in Sweden and France also said they found traces of the gene in commercially available rice. And Greenpeace said it had found traces of LL601 in rice for sale at Aldi supermarkets in Germany, prompting a nationwide recall.

How the gene spread so far is still a mystery. Rice is thought to pose a relatively low risk of cross-contamination because it self-pollinates, often before the flower even opens, lowering the likelihood that wind or insects could spread GM pollen. Steve Linscombe, a rice breeder at Louisiana State University (LSU) in Baton Rouge, where some of the test plots were grown, says they strictly followed USDA standards, exceeding the minimum requirements for buffer zones between the test plots and conventional rice. However, the university did say it found "traces of genetic material" from LL601 in samples

Fertile questions. Scientists are trying to trace how an experimental strain of genetically modified rice spread to rice sold in the U.S. and Europe.

CREDIT: KAREN TAM/*WINSTON-SALEM JOURNAL*/AP PHOTO

of foundation seed rice grown at LSU in 2003 for the widely grown Cheniere variety. Foundation seed is the original stock of a commercially available variety. It is distributed to seed-producing farmers, who then plant it to grow seed rice that is sold nationwide. Linscombe says the university is working with USDA to determine how the LL601 gene could have entered the Cheniere seed stocks.

Doug Gurian-Sherman of the Center for Food Safety in Washington, D.C., says regulations designed to limit the spread of introduced genes should require more extensive testing of such seed stocks. The possibility of contamination "needs to be taken seriously," he says. Ellstrand says that a careful investigation of what led to the spread will be crucial for scientists planning field trials of GM plants that contain more sensitive genes, such as those for pharmaceuticals or industrial products.

–GRETCHEN VOGEL

BIOETHICS

Researchers Attack Newspaper Probe of Trials

More than 100 clinical researchers have published a scathing critique of a lengthy newspaper article, which had suggested that a National Institutes of Health (NIH) researcher designed two drug trials to favor the products of company sponsors. The researcher, Thomas Walsh, an expert on treatment of infections in patients with cancer and immune deficiencies, was also a target of a congressional panel last week looking into how NIH disciplined scientists who broke rules on consulting with drug companies.

The lead author says the unusual publication is partly a response to a wave of recent media coverage suggesting that clinical trials are "rigged." "This sensationalism is hurting the process of drug approval and is hurting patients," says Elias Anaissie of the University of Arkansas for Medical Sciences in Little Rock, who with 108 co-authors published the online commentary in *Clinical Infectious Diseases* last week.

In the 5700-word report on 16 July, the *Los Angeles Times* detailed Walsh's role in leading clinical trials of two new antifungal drugs. The report suggested that doses of the older drugs being compared were too low. It also questioned whether a federal employee should have presented the companies' data to the U.S. Food and Drug Administration.

The 13 September journal commentary accuses the newspaper of "unfairly malign[ing]" Walsh and "fear-mongering" by suggesting that "the entire process of drug development … is corrupt." The researchers, 10 of whom co-authored trial publications, say the doses used were the standard of care. A footnote to the commentary describes many of the writers' extensive ties to drug companies. "You can't work in this field and not work with pharma. It's impossible," says Anaissie.

A House Commerce subcommittee last week grilled federal officials about why Walsh and another researcher who broke consulting rules are still working at NIH (*Science*NOW, 13 September, sciencenow.sciencemag.org/cgi/content/full/2006/913/1). Last year, NIH found Walsh guilty of "serious misconduct" for accepting about $100,000 from 25 drug companies without seeking permission or reporting the income. But the congressional panel is not pursuing Walsh's role in the two trials, says spokesperson Kevin Schweers. It is "following the money, not the science," he says.

–JOCELYN KAISER

CLINICAL TRIALS

A Shot of Bone Marrow Can Help the Heart

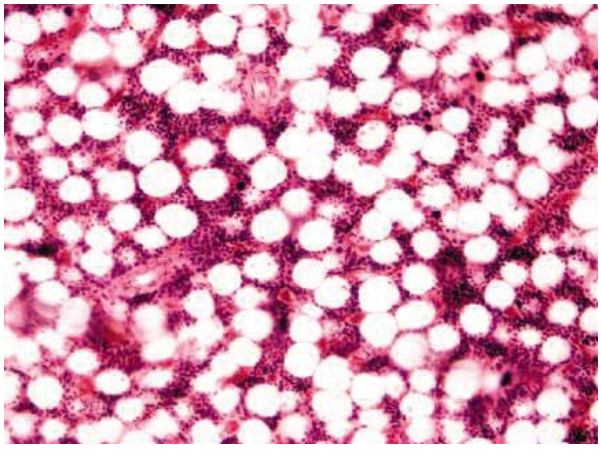
For people lucky enough to pull through after a massive heart attack, survival is only the beginning. Many patients later develop heart failure or suffer additional heart attacks. In a bid to improve the health of heart attack survivors, scientists in Germany and Norway have tested a drastic experimental treatment: shooting bone marrow straight into the heart. The hope is that bone marrow cells, which include certain adult stem cells, may trigger formation of new blood vessels, new heart muscle, or send signals to damaged muscle to repair itself.

Results of the three trials, which all appear in this week's *New England Journal of Medicine*, are contradictory, however, revealing benefit in some cases and not others and reflecting both promise and lingering confusion in this nascent field (*Science*, 9 April 2004, p. 192). One important question is whether certain types of bone marrow cells are more effective in cardiac repair than others—and how they might be working to help the heart. "That's the $100 million question," says Andreas Zeiher, a cardiologist at the University of Frankfurt in Germany, who led two of the trials.

Zeiher and his colleagues ran one trial in 204 volunteers who had had a heart attack within the previous week and another in 75 whose heart attack had hit, on average, more than 6 years before. In the trial of new survivors, Zeiher's group offered half of the participants an infusion of their own bone marrow into the affected artery. The others received a placebo injection. The study looked at left ventricular ejection fraction, a measure of the heart's pumping capacity. Four months after treatment, the bone marrow group's ejection fraction was 2.5% better than the placebo group's—a small difference, Zeiher admits, but one bolstered by the suggestion that after a year, the treated volunteers were healthier. Two had died and none had had another heart attack, compared with six deaths and five heart attacks in the placebo group.

The study wasn't big enough to assess definitively whether these differences were real or occurred by chance. Still, "it shows that there's a therapeutic window that is much larger than we previously thought," says Douglas Losordo, who is moving from Tufts University in Boston to direct the Feinberg Cardiovascular Research Institute at Northwestern University in Chicago, Illinois. A favored slogan of cardiologists—"time is muscle"—comes from the idea that doctors must treat patients within hours of a heart attack in order to have a measurable effect on their cardiac function, says Losordo.

Zeiher's second trial, of individuals who, on average, had had a heart attack more than 6 years earlier, was smaller and produced results that were less clear-cut. Those given their own bone marrow were better off, with an ejection fraction that exceeded the control group's by 4.1%. Another group who received similar types of cells culled from their blood, instead of their bone marrow, experienced no benefit. Losordo, who is running a clinical trial that also selects cells from the blood, believes the blood portion failed because too few cells were administered.



Recipe for repair? Two studies find that bone marrow (*above*) improves cardiac function, but a third reported no effect.

CREDIT: BCC MICROIMAGING/VISUALS UNLIMITED/GETTY IMAGES

"A lot of people [may] look at these trials and say, 'The benefit seems to be pretty small,' " says Richard Cannon, clinical director for the division of intramural research at the National Heart, Lung, and Blood Institute in Bethesda, Maryland. Giving him pause, too, is the Norwegian study, led by cardiologists Kolbjørn Forfang and Ketil Lunde of the Rikshospitalet ▶

A Lunar To-Do List

NASA asked the scientific community what science it should do when it returns humans to the moon, and this week it got a quick answer: the same things scientists have been saying all along. A report released by the National Research Council draws on previous recommendations for an ambitious, moon-girdling effort to understand the origins of such rocky bodies.

NASA wanted prioritized research objectives for the robotic orbiters and landers that will primarily act as scouts and for the astronauts who will explore the moon's surface, initially in beefed-up Apollo-style missions. The study committee, headed by retired Aerospace Corporation executive George Paulikas, calls programs for lunar data analysis its top priority. Next is exploration of the South Pole–Aitken basin, an impact scar mostly on the moon's back side. Then comes an instrument network for probing the interior, followed by rock sample returns, scientifically selected landing sites, and analysis of any icy polar deposits.

The targeted "science hasn't changed," says committee member Carlé Pieters of Brown University in Providence, Rhode Island. The objectives remain the same as when robots were going to do all the exploration.

–RICHARD A. KERR

Respect for Authority

A House panel this week was expected to grant new budget authority to the director of the National Institutes of Health (NIH)—and endorse a 5% annual raise for the $28.6 billion agency.

The so-called reauthorization bill, introduced by Representative Joe Barton (R–TX), chair of the House Energy and Commerce Committee, doesn't give the biomedical giant a dime. And Congress isn't expected to complete action on the measure until sometime next year. But lobbyists like its contents, including its support for solid spending boosts through 2009.

Barton has eliminated an earlier controversial proposal that would have divided NIH's budget for its 27 institutes and centers into two pots—one disease-oriented and the other "science-enabling" (*Science*, 22 July 2005, p. 545). The bill would hold steady the total number of institutes and create a division to plan trans-NIH initiatives through pooled funds that would eventually encompass 5% of NIH's budget. "It's a really positive bill," says Jon Retzlaff of the Federation of American Societies for Experimental Biology in Bethesda, Maryland.

–JOCELYN KAISER

University Hospital in Oslo. "We found no difference" between 47 individuals given bone marrow roughly 6 days after a heart attack and 50 others in a control group, says Lunde. The study was not designed to pick up a disparity of less than 5% in ejection fraction, he notes, a difference he feels is necessary to recommend a treatment as invasive as this one. In all three studies, the treatment appeared safe.

"We need to move to the next level [in trials], and the next level has to be survival and prevention of heart failure," says Cannon. Another key question, says Losordo, is what type of bone marrow cells to use. "There's abundant evidence … that not all bone marrow cells are created equal," he notes. The German team had many more cells that boasted the surface marker CD34, the same one Losordo and others use to mark "stemness." Whereas Lunde has no plans to pursue the therapy further, Zeiher is looking toward a trial of 1200 patients. "I don't think the data's strong enough to say we should start doing this to everybody," says Joshua Hare, a cardiologist at Johns Hopkins University in Baltimore, Maryland. "But it clearly substantiates that we should move ahead."

–JENNIFER COUZIN

PALEOANTHROPOLOGY

Lucy's 'Child' Offers Rare Glimpse of an Ancient Toddler

After a long, hard labor, a young Ethiopian has delivered a tiny bundle to the paleoanthropological community: the fragile skeleton of a 3-year-old girl buried about 3.3 million years ago in a flood in Ethiopia. At least half of the tot's fragile skeleton is preserved, including the skull with both jawbones attached. It is the "most complete partial skeleton of an early juvenile hominid ever discovered," exults its discoverer, Ethiopian paleoanthropologist Zeresenay Alemseged of the Max Planck Institute for Evolutionary Anthropology in Leipzig, Germany.

After working for 5 years with dental tools to painstakingly remove cementlike sandstone from the skeleton, Alemseged uncovered enough of the hominid to offer a viewing of it in this week's issue of *Nature*. Analysis of the freshly exposed skeleton has just begun and the bones of the upper body are still stuck together, but the remains promise to give researchers their first glimpse of a juvenile hominid from head to toe, as well as rare insight into ancient hominid growth and development. Although there are older jaw fragments of children, there are only two other child's skulls dating to more than half a million years ago, including a deformed skull of the same species. "There hasn't ever been a fossil of that antiquity with so many winning cards," says paleoanthropologist Bernard Wood of George Washington University in Washington, D.C.

Alemseged was a postdoctoral researcher at Arizona State University's Institute of Human Origins in Tempe in December 2000 when he and Ethiopian antiquities officer Tilahun Gebreselassie found part of the skull protruding from the ground in Dikika, Ethiopia. Over the next 4 years, Alemseged and his colleagues slowly extracted the petite skeleton. It had been buried by floodwaters about 10 kilometers from the resting place of the famous 3.1-million-year-old partial skeleton called Lucy, at Hadar, and turns out to be an earlier member of her species, *Australopithecus afarensis*.

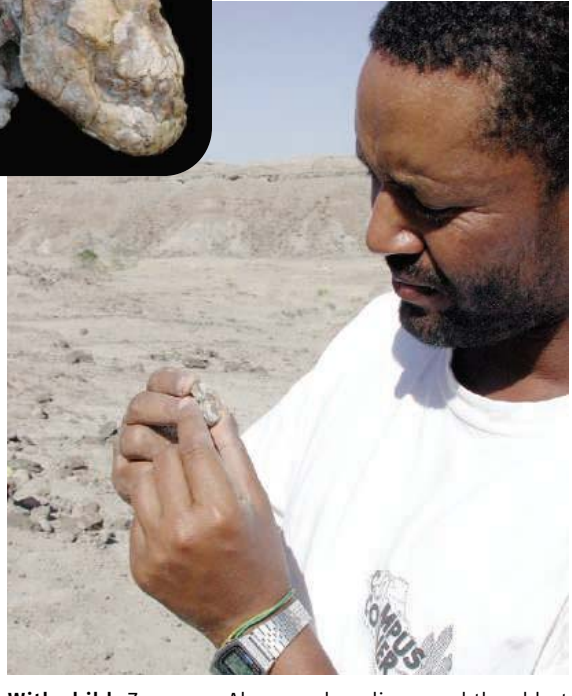
As in adults of this species, the child has a mix of primitive, apelike traits and more modern traits such as ankle and knee bones adapted for upright walking. The remarkably complete skeleton includes all the teeth, which were used to determine age and sex, plus rare bones such as shoulder blades and an apelike hyoid bone, only the second ancient voice box bone found. The shoulder bones looked so much like those of a young gorilla that it was a "shock," says co-author Fred Spoor of University College London. The shoulder blades and long, curving fingers suggest to Alemseged that *A. afarensis* might have been adapted for climbing in trees. This reignites a long-standing debate over whether this upright species still spent much time in the trees or merely retained ancient features it no longer used, like wisdom teeth in humans.

Indeed, Carol V. Ward of the University of Missouri, Columbia, an expert on hominid postcranial bones, says that photos of the shoulder bone did not look so apelike to her. And Tim White of the University of California, Berkeley, warns that it is premature to draw conclusions about the fossils' development and locomotion, because the team hasn't yet finished cleaning the rock from the bones.

Nonetheless, Ward and others predicted that the skeleton would offer an "exciting opportunity" to look at skeletal development and how proportions of limbs and body parts developed in relation to each other and to other fossils. A comparison with the famous 4-year-old Taung child skull of *A. africanus*, whose species lived 1 million to 3 million years ago in South Africa, might also reveal differences in development.

The Dikika girl had a small, chimp-sized brain, as expected for her species. But her brain might have grown at a slower rate than in apes—perhaps resembling the slow rate found in humans, says Alemseged. This may be a hint of a more humanlike pattern of development or just a poor diet. Regardless, says Alemseged, at the very least, "now we can say: This is what a baby *A. afarensis* looks like."

–ANN GIBBONS



With child. Zeresenay Alemseged co-discovered the oldest skeleton of a child in the badlands of Ethiopia.

CREDITS: DIKIKA RESEARCH PROJECT; (INSET) ZERESENAY ALEMSEGED

OCEAN SCIENCE

Creatures Great and Small Are Stirring the Ocean

Explaining the forces that mix things up in the ocean has always been the province of the physical oceanographers. It seemed obvious that physics governs how the winds and tides drive the waters and thus how deep, cold, nutrient-rich seawater is mixed toward the surface. Marine life was clearly just along for the ride.

But recent evidence suggests that marine life may itself be helping stir the ocean, from local to global scales. "My initial reaction was, 'Preposterous,' " says physical oceanographer Carl Wunsch of the Massachusetts Institute of Technology in Cambridge. "Then you look at the numbers, and it's not so preposterous. The order-of-magnitude numbers suggest it's worth talking about." And the implications could be huge. Overfishing of the big whales, for example, could be changing global climate.

On page 1768, physical and biological oceanographers report that at least one rather small marine creature does indeed stir the ocean, if only a small part of it. Prodded by theoretical claims of substantial biomixing on a global scale, physical oceanographers Eric Kunze, Richard Dewey, and Kevin Bartlett and biological oceanographers John Dower and Ian Beveridge, all at the University of Victoria in Canada, went to Saanich Inlet on the British Columbia coast to take a close look. This fjord's water is stratified, and turbulence there is as low as in the deep, open ocean.

Saanich Inlet is also home to myriad 1- to 2-centimeter-long, shrimplike creatures called krill. They loiter in the murky depths by day and at night swim up toward the surface under the cover of darkness to feed. So Kunze and his colleagues lowered an instrument package into the path of the migrating krill. The measured turbulence shot up by three to four orders of magnitude for 10 to 15 minutes as the krill passed.

"I'm pleased it isn't all just theory," says biological oceanographer Mark Huntley of the University of Hawaii, Kaneohe. He and biological oceanographer Meng Zhou of the University of Massachusetts, Boston, had calculated how much mixing a variety of critters—from bacteria to blue whales—might be causing, based on what is known of their size and swimming habits. By Huntley and Zhou's calculations, the turbulent mixing of schooling animals from krill and anchovies to whales "is equivalent to a pretty sustained storm at a local level," says Huntley.

Oceanic agitator. This 2-centimeter krill, *Euphausia pacifica*, can mix the sea if it teams up with thousands of its kind.

CREDIT: MOIRA GALBRAITH

Huntley imagines that schools of krill in the stratified Southern Ocean around Antarctica could be stirring water upward with each daily vertical migration, locally replenishing the nutrients depleted by the photosynthesizing phytoplankton. In effect, the swarming krill could be fertilizing the shallow sea, which would boost the production of phytoplankton that are eaten by small zooplankton that feed the krill.

Grander speculations will appear in the upcoming *Journal of Marine Research* (July 2006 issue). Physical and biological oceanographers led by William Dewar of Florida State University in Tallahassee calculated how much energy phytoplankton store in new organic matter each year: about 63×10^{12} watts (63 terawatts, or TW). Perhaps something like 1% of that, or almost 1 TW, may go into swimming motions that stir ocean waters, they estimate from expected energy losses and from the amount of oxygen consumed in the ocean.

A TW of biomixing would be a lot. In 1998, Walter Munk of the Scripps Institution of Oceanography in San Diego, California, and Wunsch estimated that 2 TW of mixing is required to mix deep, cold waters to the surface. That completes the "conveyor belt" circulation of the world ocean, which is vital to the climate system. Dewar and his colleagues speculate that the decimation of stocks of big fish and whales over the past couple of centuries could have removed enough biomixing to have an effect on climate.

–RICHARD A. KERR

Prized Science

This year's Lasker awards for medical science span generations as well as disciplines. University of Pennsylvania psychiatrist Aaron Beck won an award for developing cognitive therapy, and pioneering cell biologist Joseph Gall, inventor of the gene-finding technique called in situ hybridization, was honored for a career of achievement. Researchers Elizabeth Blackburn (University of California, San Francisco), Carol Greider (Johns Hopkins University School of Medicine), and Jack Szostak (Harvard) shared an award for their discovery in the 1970s and 1980s of the enzyme that makes the ends of chromosomes, which has led to potential treatments for cancer and age-related ailments. "We had no idea this was going to have medical implications," says Greider, calling the award a testament to "curiosity-driven" science.

–ELI KINTISCH

Exports: A Matter of Great Import

The U.S. Commerce Department has asked a panel of experts to examine whether policies on limiting access to sensitive information and technologies should be reviewed. The move comes a year after the department proposed tougher rules on so-called deemed exports; universities and companies argued that the regime would hinder research. That proposal was shelved in May (*Science*, 19 May, p. 985).

The 12-member panel will be headed by Norman Augustine, the former Lockheed Martin head, and Robert Gates, current president of Texas A&M University. Both served on a National Academies committee on national competitiveness that last year called for relaxing deemed-export rules.

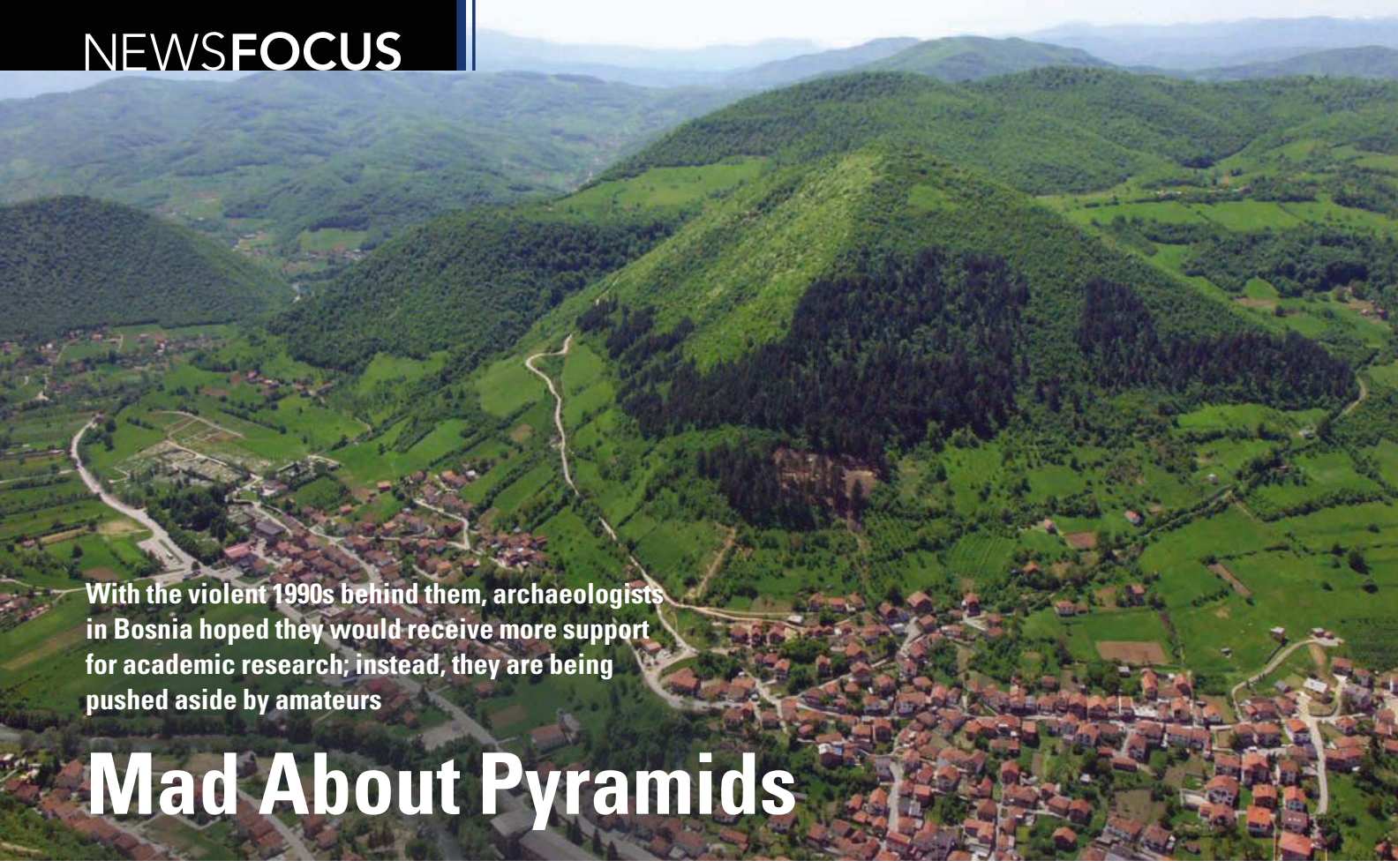
–YUDHIJIT BHATTACHARJEE

Greenhouse Report

Cutting greenhouse emissions requires both research on new technologies as well as market limits on carbon usage, says the Congressional Budget Office in a new report.

The report implicitly criticizes the Bush Administration's emphasis on energy research, arguing that "relying exclusively on R&D funding in the near term … would increase the overall cost of reducing emissions in the long run." Senator Jeff Bingaman (D–NM), who requested the study with independent James Jeffords (VT), says the work validates his call for carbon limits. The Competitive Enterprise Institute's Chris Horner thinks that strategy is unwise, pointing out that high gas prices in Europe, for example, have not led to lower emissions.

–ELI KINTISCH



With the violent 1990s behind them, archaeologists in Bosnia hoped they would receive more support for academic research; instead, they are being pushed aside by amateurs

Mad About Pyramids

SARAJEVO—It should have been a great day for Balkan archaeology. For the first time since the bloody civil war, experts from all corners of ethnically divided Bosnia gathered for an impromptu meeting at the National Museum. Television crews were waiting outside for interviews. Foreign scientists were on hand, too—including the president of the European Association of Archaeologists, Anthony Harding of Exeter University in the U.K.

But the mood was one of deep frustration. The journalists weren't interested in the scientists' plans for restarting international collaborations. Nor did they want to hear about rebuilding the ailing university curriculum, or saving the country's archaeological assets from neglect and looting. "They only want to hear about one thing," says Zilka Kujundzic-Vejzagic, the museum's expert in prehistoric archaeology, who organized that 9 June meeting: "pyramids, pyramids, pyramids."

The "pyramids" in question are 30 kilometers northwest of Sarajevo near the town of Visoko. A Bosnian businessman named Semir Osmanagic, who runs a construction company in Houston, Texas, announced last year that a 360-meter-tall hill that looms over Visoko is in fact a buried pyramid built, he claims, by an unknown civilization 12,000 years ago. He has dubbed it the Pyramid of the Sun. With the help of volunteers, Osmanagic has uncovered stone blocks beneath the hill's surface and a system of tunnels, which he says are like those of the pyramids in Egypt. Osmanagic has proposed that two smaller hills nearby are part of the same "pyramidal complex."

That vision is not shared by any of a half-dozen archaeologists and geologists who spoke to *Science* after visiting Visoko. The truth is plain, says Stjepan Coric, a Bosnian geologist at the University of Vienna, Austria, who was invited by Osmanagic to examine the site: The stone slabs are nothing more than fractured chunks of sediment called breccia, the remains of a 7-million-year-old lakebed that was thrust up by natural forces. "This is what gives the mound its angular shape," Coric says. As for the tunnels, "if they were made by humans, without establishing their age, I would assume they are part of an old mine." Harding's verdict: "It's just a hill."

But this humdrum assessment has been swept aside by a pyramid-mania that has gripped the media. Osmanagic, aided by a publicist and an Indiana Jones–style hat, is widely depicted as a maverick bravely pursuing his unorthodox hypothesis. Even the BBC contributed a wide-eyed report in April. The Bosnian public and politicians have fallen deeply under his spell. Archaeologists are concerned that funding for real research projects is being drained away to support Osmanagic's "Pyramid of the Sun Foundation," and those who voice dissent are receiving hate mail. "To believe in the pyramids has become synonymous with patriotism," says Kujundzic-Vejzagic. Worse than that, some archaeologists say, Osmanagic is starting to dig up the remains of unstudied human occupation, possibly a long-sought medieval town. "Pyramid-mania" will probably be short-lived, says Harding, but it would be "tragic" if it damaged "real archaeological material."

Picking up the pieces

"Sarajevo was a real center of excellence" for archaeology before the war broke out in 1992,

CREDIT: MARIO GERUSSI

Human design? The hill that looms over Visoko resembles a pyramid.

says Harding. But during 4 years of nonstop shelling, "we nearly lost everyone and everything," says Kujundzic-Vejzagic, who fled to Croatia a year into the conflict.

Archaeological sites were used as defensive positions in fierce battles, and shattered windows left the museum vulnerable to winter weather and animals. The timing could not have been worse, says Preston Miracle, an archaeologist at the University of Cambridge, U.K., who has worked in the region for 2 decades. Just before the war, he says, "the senior generation of Bosnian prehistorians all died," and the generation in line to replace them scattered.

Ten years on, the community still has not recovered, "but at least it is clear what needs to be done to get us back in shape," says Kujundzic-Vejzagic, who returned in 1998 and has remained at her post in Sarajevo. The first priority is "to assess and protect" the endangered archaeological riches in the country, now known as Bosnia and Herzegovina. This roughly Switzerland-sized territory has been continuously occupied all the way back to the last Ice Age and beyond.

Little is known about the first Slavic tribes that arrived some 1500 years ago, says Kujundzic-Vejzagic. Even less is known about the people who preceded them, the Illyrians, who held sway from around 1300 B.C.E. until the Romans took over. Learning more about their interactions with neighboring cultures, especially the Greeks, would shed light on the technological revolutions that changed Bronze and Iron Age Europe. "All of these settlements and graves are just waiting to be studied," she says, although "we'd do better to leave everything in the ground" until resources are secured to protect against weather and looters.

Deeper in time, fundamental questions about Neolithic society have sustained one of the few remaining international collaborations in Bosnia. Over the past 4 years, a team led by Kujundzic-Vejzagic and Johannes Müller, an archaeologist at the University of Kiel, Germany, has been exploring a site near the town of Okoliste, 7 km away from the pyramid hunt. It has been identified as part of the Butmir culture, a source of richly decorated pottery and intricate statuettes discovered in 1893. Research on these artifacts and related 7000-year-old dwelling sites could help answer one of the central questions of prehistoric archaeology, says Müller: "How and why did we go from simple, egalitarian societies of small settlements to complex, hierarchical societies with big, dense settlements?"

Buried in the soil near Okoliste are the remains of the largest Neolithic settlement ever found in Europe: between 200 and 300 houses protected by a ring of three trenches and a raised bank. "I was astonished when I realized that this defended area alone could have been home to as many as 3000 people," Müller says. Settlements from contemporary Neolithic cultures in Europe were occupied by no more than 300.

Another research team, led by Miracle and Tonko Rajkovaca, a Bosnian archaeologist also at Cambridge, has just begun looking for traces of even earlier human occupation in northern Bosnia; the area is thought to be one of the last refuges of the Neandertals. "Despite the richness of this record," says Miracle, the region "remains poorly known and understood."

Big dig. Semir Osmanagic (in hat) and Ivica Šarić, Sarajevo's minister of culture, with volunteer excavators at the "pyramid" site. Archaeologists worry that valuable material may be stripped away.

With relatively untapped heritage resources, academic archaeologists say, the Bosnian government should be trying to help in any way possible. But instead, many researchers feel that the country is turning against them.

When hills become pyramids

If you stand in the right place in Visoko, the largest of the nearby hills almost looks like a pyramid. At least, two of its sides are more or less flat, although the rest is lumpy. During a tour of the site by *Science* in June, freshly dug earthen stairs led up the slope through the trees, slick with rain. Along the way up, broad patches of soil had been cut away and roped off with yellow tape. A pair of local guides pointed to the exposed crust of fractured rock and explained, "This is the side of the Pyramid of the Sun." And pointing to two smaller hills across the valley: "That is the pyramid of the dragon, and that one is the pyramid of the moon."

Osmanagic, who came up with the hills' mythical names, says he became convinced in April 2005 that they are buried pyramids, based on their shape and position. Osmanagic is in love with pyramids. He says he has studied "hundreds" of them around the world—including the Mayan pyramids, which in his view were located and built with "vibrational" technology inherited from the lost civilizations of "Atlantis and Lemuria."

He says he has sought the help of experts to make "serious scientific argumentation." One of the first was Amer Smailbegovic, a geophysicist who runs a surveying company and teaches at the International University of Sarajevo. "I noticed that the area has a peculiar triangular-sided feature you don't see too often in a temperate environment," says Smailbegovic, who analyzed satellite imagery for Osmanagic. Thermal and radar imaging also made the hill seem "out of the ordinary," he says. So Smailbegovic wrote to Osmanagic that "there are anomalies present in your area of interest, and you may have something there. I suggest you find yourself an archaeologist and geologist to help you validate the area." But "the next thing I know," Smailbegovic says, "there was a

CREDIT: MARIO GERUSSI

headline in the Bosnian papers: Satellite imagery confirms Osmanagic's discovery of pyramids in Bosnia." This would prove to be the start of a barrage of "sensationalism," he says.

Osmanagic says he invested $20,000 of his own money to hire dozens of people, including a public relations manager, and established a tax-exempt foundation to pay them. He also placed an advertisement in the listings of the Archaeological Institute of America for someone who could do Paleolithic fieldwork. Among those who responded was Royce Richards, an archaeologist who works for the Australian government as a heritage officer in Adelaide.

In January, "things got very strange," says Richards. In newspaper articles around the world, he was named as one of the main "expert advisers" on an international dig that has discovered "evidence of Bosnian pyramids." Osmanagic's foundation Web site had included Richards in the "advisory committee of experts," even though he never visited Bosnia nor confirmed that he would participate. Other academics say they were listed although they had never asked to be involved. One of them, Bruce Hitchner, head of the archaeology program at Tufts University in Medford, Massachusetts, objected when he learned that his name had been hijacked. Osmanagic has removed all the expert advisers' names from his Web site but says, "I did nothing wrong."

Smailbegovic visited Visoko in April to see the project for himself. "The situation was chaotic," he says. Osmanagic's volunteers are digging up the area, but Smailbegovic didn't see much effort directed at "answering the question of why there are geospatial anomalies in the Visoko valley." Smailbegovic and other geologists conducted their own field study of the Visoko valley in May and June. He says Osmanagic has ignored their detailed reports, which conclude that natural forces created "the majority of the landscape features" and that "meticulous archaeological work" is needed to determine whether humans had any part in it.

Osmanagic says he is doing just that, but archaeologists are outraged. "This is the equivalent of letting me, an archaeologist, perform surgery in hospitals," says Enver Imamovic of the University of Sarajevo, a former director of the National Museum. By assuming that the hills are pyramids from the very start, says Müller, "that's all he'll ever see." For example, he points out, Osmanagic's deduction of the age of the pyramids at 12,000 years old is based on nothing more than the depth of the soil over the stones that he claims are masonry. While clearing away that soil, Osmanagic's volunteers have found engraved stones and a skeleton. Imamovic worries that these may be signs of a long-sought necropolis or a lost town mentioned in Byzantine texts. Osmanagic says the skeleton "is being analyzed," but he believes it was recently interred.

Osmanagic also says he has uncovered a stone layer that is "the pyramid's face" on one of the smaller hills. A European archaeologist working in Bosnia who had a look for himself says, "There is a real wall there, but it looks to me like part of a small Middle Age rain reservoir." The archaeologist, who requested anonymity for fear of losing permission to work in the country, says he is not surprised that diggers have uncovered signs of human occupation: "People have been here for millennia." But after Osmanagic is done with Visoko, "we may never know what was really here," he says. The real archaeological material is between the surface and the bedrock, he says, "but for a pyramid-hunter, that is just dirt to strip away."

The real thing. Archaeologist Zilka Kujundzic-Vejzagic holds a pot from the Butmir culture that flourished in Bosnia 7000 years ago.

CREDIT: J. BOHANNON/SCIENCE

Osmanagic says he is aware that he is digging through layers of occupation and claims he will publish his results "in a peer-reviewed journal" in November. "But I am not interested in the approval of elite scientists. This project is for the people."

Popular archaeology

In spite of the protests from academic quarters, public and political support for Osmanagic seems to be growing. The government has granted him all the necessary permits and has even helped finance his excavations. "It is shocking" that public funds are flowing to Osmanagic instead of the country's desperate archaeologists, says Müller. But Osmanagic says that only 10% of his current budget—the total is about $300,000, he says—comes from government support, while the rest is from "private funds and corporate sponsors."

One expert says it's easy to understand why people seeking a national identity would embrace the Visoko phenomenon. "Osmanagic's pyramid fantasies are exactly what the majority of Bosnians want to hear," explains a Bosnian sociologist who spoke on condition of anonymity. There are also economic motivations. Last month, Osmanagic announced plans to build three "archaeological parks" across the country that will "rewrite world history" by revealing more evidence of Bosnia's prehistoric "supercivilization." New highways and hotels are part of the plan.

Crude as it may seem, pyramid-mania could be a boon over the long term, says Miracle: "If the energy and interest in archaeology can be redirected into Bosnia's rich heritage, then this affair would not be such a fiasco after all." But few are optimistic. Kujundzic-Vejzagic says she is on the verge of quitting. She says she's been the target of hate mail from the pro-pyramid movement; no one in government has stepped forward to defend her. If she goes, the entire Butmir project will probably fold, says Müller. "There is no other prehistoric archaeologist in the country," he says. "She is our only partner." Bosnia's other archaeologists are in an equally precarious position. A Visoko municipal official recently announced that all critics of Osmanagic's project should be denied access to research locations and have their degrees revoked.

Descending the hill back down to Visoko, a visitor wades through the friendly locals selling official "Pyramid of the Sun" T-shirts and mugs. One thing is clear: Some people will benefit from the hunt for a prehistoric Bosnian civilization. But they may not be academic archaeologists.

–JOHN BOHANNON

SCIENTIFIC WORKFORCE

Frustrations Mount Over China's High-Priced Hunt for Trophy Professors

Chinese universities bask in the glow of top-gun scientists hired on part-time deals to share their wisdom. Critics say the money could be spent more wisely

Mathematician Gang Tian did not expect a standing-room-only crowd last week when he gave a lecture at Beijing University (Beida) on the Poincaré conjecture. But not all were there for the math. Reporters and others had come for a glimpse of the man at the center of a tempest engulfing Chinese academia. Tian is a premier example of a controversial phenomenon: a Chinese-born researcher with a full-time faculty position overseas who gets paid handsomely for short working stints in his homeland.

Resentment against part-timers boiled over last July, when Shing-Tung Yau, a Harvard University mathematician and Tian's former mentor, dismissed the "majority" of Beida's overseas recruits as "*jiade*," or "fakes," in comments in the Chinese magazine *Nanfang Renwu Zhoukan*. Beida officials fired off a series of rebuttals in which they termed Yau's remarks "irresponsible" and a "distortion of facts" and rattled off achievements—papers in prestigious journals and patents, for instance—by talent returned from overseas.

The university's attempts at damage control, however, only intensified debate about professors such as Tian, who has been listed among Beida's full-time faculty for several years. Beida nominated Tian to membership in the Chinese Academy of Sciences (CAS), an honor reserved for scientists who expend at least half their effort in China. Thanks to Beida's backing, Tian—who was then also listed as a full-time professor at the Massachusetts Institute of Technology (MIT)—was elected in 2001 by a margin of one vote. Last spring, Tian left MIT to become a full-time professor at Princeton University.

After the Poincaré lecture, reporters pressed Tian about his employment status in China. He said that he now spends more than 4 months a year at Beida and "hopes to be a full-time professor later on," perhaps after Beida builds a $13 million international institute of mathematics, which Tian will direct.

A call for oversight. Shigang He thinks China's funding agencies should hold part-time professors to their contractual commitments.

CREDIT: COURTESY OF SHIGANG HE

Some proponents consider part-time academic appointments a critical means of stanching China's loss of scientific talent. Universities and government agencies are boosting quotas for part-timers and upping the ante to entice more top guns to return. Several universities have created "million-yuan professorships" with stratospheric—for China—annual salaries equivalent to $125,000. Most returnees are midcareer scientists who accept more modest offers (see sidebar on p. 1722).

Critics, however, contend that part-timers often are less important as professors than as tools in the battle for prestige and resources. Yau claims that researchers who parachute in can hardly contribute in a substantive way to China's scientific development. But the trend seems almost unstoppable, says Shigang He, a neuroscientist currently at CAS's Institute of Biophysics in Beijing: "I don't think universities will really seriously control this, because they benefit."

Offers too good to refuse?

Overseas academics began returning to China in the late 1990s, drawn by programs to woo talented scientists back (*Science*, 21 January 2000, p. 417). The Ministry of Education's Changjiang Scholars Program and CAS's One-Hundred-Talent Plan intended initially to recruit people to work at least 9 months a year—essentially full-time—in China. But top-notch researchers who signed up wanted to help their homeland and keep their jobs overseas: "If you have a tenured professorship [in the United States], it does not make sense to give up the position," says Jun Liu, a statistician at Harvard.

The education ministry quickly took a new tack, creating a category of part-time Changjiang scholars: *jiangzuo*, or lecture chairs, for associate professors or higher. They are required to spend no fewer than 3 months—or two, "under special circumstances"—in China. But universities eager to attract stars are willing to make exceptions. Ying Xu, a bioinformatics researcher at the University of Georgia, Athens, says he turned down a couple of invitations to apply for a *jiangzuo* post, citing time constraints. University officials have told him that a 3-month commitment could be met by arriving at the end of the first month and leaving at the beginning of the third—but Xu says "his conscience did not allow" him to play that game. (Such overtures, other scientists say, are typical.) Xu chose instead to organize a weeklong symposium in China each summer.

Other part-timers say they are unaware of a time requirement. Liu accepted a *jiangzuo* post at Beida in 2002, but he acknowledges that he spends only about 1 month a year in China. Gary Becker, a Nobel laureate in economics at the University of Chicago in Illinois who recently joined Beida as a Changjiang *jiangzuo*, says, "What I will do is not precise; it will be mainly up to me."

Incentive programs have stirred controversy before. CAS began a crackdown after an open letter in 2003 publicized one extreme case of a full-time researcher then at the University of Wisconsin who held grants from three programs and fulfilled a pair of 9-month and one 6-month commitments concurrently. According to CAS's Li Hefeng, the academy so far has canceled the awards of 166 recipients (out of 1005 overseas recruits) and demanded the money back.

Despite allegations that the system is rife with cheating, universities covet part-timers and have lobbied for an expansion of the programs. In 2004, the education ministry raised its annual quota of *jiangzuo* from 10 to 100. Last year, Beida for the first time appointed

Many Overseas Chinese Researchers Find Coming Home a Revelation

SHANGHAI—"When I left China to study abroad, I thought I had left China for good," says neuroscientist Shigang He. Yet, after earning his Ph.D. and landing a permanent research position in Australia, He started having second thoughts. A visit to a Chinese institute astounded him. Labs were bulging with new equipment and feverish with activity. And funding for individual researchers was nearly on a par with his in Australia. He made several trips back to China, he says, "to make sure I wasn't deluded." Then he did something once unthinkable for a Chinese scientist established abroad: He resigned from the University of Queensland, sold his house in Brisbane, and joined the Institute of Neuroscience in Shanghai, a part of the Chinese Academy of Sciences (CAS).

"I've never regretted it," says He, who is now with the CAS Institute of Biophysics in Beijing. "For my research, and personally, it was a good decision."

He's not alone. Although numbers are hard to come by, repatriated scientists are multiplying. Officials at the Institute of Health Sciences, a part of CAS's Shanghai Institutes for Biological Sciences (SIBS), say a third of the two dozen primary investigators who have joined the institute since its founding 4 years ago had given up permanent jobs overseas. "It is definitely a new trend, not only at SIBS but throughout China," says SIBS President Gang Pei.

Those returning to their roots say the trend indicates how far Chinese science has come in catching up with the West. "It is no longer true that a faculty position in China is less competitive than one in the U.S.," asserts Jianmin Zhou, a molecular plant biologist who left an associate professorship at Kansas State University, Manhattan, for a position at the National Institute of Biological Sciences in Beijing. In China, midcareer returnees bridge a gap between young scientists trained abroad and high-profile veterans who spend a few months a year in China as advisers. "These midcareer people help China" with their experience and administrative skills, says Pei.

Making a choice. Population geneticist Li Jin gave up a U.S. tenured professorship to become dean of the School of Life Sciences at Fudan University.

The returnees so far, however, are not superstars. Few "are from first-tier universities and/or doing first-rate work," says Li Jin, a population geneticist who relinquished a full professorship at the University of Cincinnati to become dean of life sciences at Fudan University in Shanghai. And returnees spurn offers from any but the top institutions in cosmopolitan Beijing and Shanghai.

Deciding to come home usually starts with a realization of how quickly more Changjiang part-time (11) than full-time professors (10). Many universities have set up their own programs for illustrious part-timers—"Nobel laureates" and "internationally famous scholars," as Zhejiang University's announcement puts it. Whereas Zhejiang is still hoping to snare a Nobel laureate, Beida, in rebutting Yau, touted three among its *jiangzuo* ranks: "One can well imagine their contributions to education and research," the university stated.

A fair compromise?

Many academics feel that the prestige that comes with hiring part-timers is superficial. "Some high-profile papers appear to come from China, even though the science didn't really take root [there]," says Mu-Ming Poo, a neuroscientist at the University of California, Berkeley. Chinese universities turn a blind eye to absentee professors as long as they list their Chinese affiliation on papers, adds He.

Indeed, the number of publications with Chinese authors listing multiple affiliations is on the rise. For example, Zhong Lin Wang, a nanotechnology researcher at Georgia Institute of Technology in Atlanta, has three affiliations on recent papers in *Science*: Georgia Tech, Beida, and the National Center for Nanoscience and Technology in Beijing. Wang is part-time director at both Chinese institutions, which hailed his publications on their Web sites. Wang acknowledges that the work was done solely at Georgia Tech. Similar cases abound.

At the same time, some part-timers downplay their moonlighting. Zhensu She, a mathematician at the University of California, Los Angeles, lists in his CV on UCLA's Web site his full-time Changjiang professorship at Beida as an "award" in 1999—it was a 5-year contract—and does not mention that he is director of the Key State Laboratory of Turbulence and Complex Systems and deputy director of the Center of Theoretical Biology, both at Beida.

UCLA policy states that "compensated teaching or research at another institution while employed as a full-time faculty member" requires "prior written approval of only the Chancellor or Executive Vice Chancellor." As *Science* went to press, UCLA had not clarified whether She or seven other faculty members with similar positions in China obtained such approval. She did not respond to requests for an interview; a source in UCLA's math department says he is on sabbatical.

Teaming up—or outsourcing?

To many Chinese scientists, the bottom line is not how much time is spent on Chinese soil but whether one contributes to the country's science. Poo helped create the Institute of Neuroscience (ION) in Shanghai in 1999 and since then has been its part-time director. He views his role as enabling young Chinese scientists to gain international recognition based on their own projects and publications. Although Poo spent about 80 days in Shanghai last year, and ION covers his expenses, he does not receive an ION salary. "I do not have any problems with people like Mu-Ming

	Tepin (full-time: 9 months)							*Jiangzuo* (part-time: 2–3 months)						
University	**1999**	**2000**	**2001**	**2002**	**2003**	**2004**	**2005**	**1999**	**2000**	**2001**	**2002**	**2003**	**2004**	**2005**
Beijing	6	8	9	15	9	11	10	3	4	3	1	1	8	11
Qinghua	5	14	6	10	4	7	7	0	3	1	4	1	7	9
Fudan	6	7	3	4	5	7	8	1	1	0	0	1	5	6
Nanjing	2	6	3	8	5	6	4	0	0	0	0	1	5	2
Zhejiang	2	2	3	9	7	4	6	1	1	0	0	0	3	0
Shanghai Jiao Tong	3	5	7	7	1	4	4	1	0	0	0	0	2	6
(all universities)	66	112	97	135	84	111	101	6	10	10	7	10	79	89

Buying spree. Top Chinese universities are sharply increasing their ranks of part-time researchers from overseas, even as numbers of full-time returnees hold steady.

CREDITS (TOP) COURTESY OF LI JIN; SOURCE (BOTTOM): MINISTRY OF EDUCATION

the research landscape in China is improving. "Support for research by most agencies in China doubled this year," says Jin. "The pressure for getting grants [abroad] is one of the major reasons that drives people back to China." The funding now offered a new scientist to set up a lab in China roughly matches that of a U.S. university, adds He. Individual grants may be smaller, but money goes further in China too. Scientists also rave about the quality of students. "Many Chinese faculty … have better students than their peers in the U.S.," says Zhou. Part of the appeal is helping shape China's science future. Guo-Tong Xu, once an assistant professor at the University of North Texas Health Science Center in Fort Worth and now deputy director of SIBS's Institute of Health Sciences, boasts that his institute is the first in China dedicated to translational research, bridging the gap between basic and clinical research.

Some scientists return to pursue opportunities that are illegal or not encouraged in the United States and other countries because of ethical concerns. Hui Zhen Sheng returned in 1999 after 10 years at the U.S. National Institutes of Health when the Shanghai government made an offer "too good to decline": to fund a stem cell lab for her at Shanghai Second Medical University. Sheng is working on therapeutic cloning of human embryonic stem cells using animal eggs.

Similarly, stem cell research brought Hongkui Deng back. Deng left China in 1989 to study immunology at the University of California, Los Angeles, and later became research director for ViaCell Inc., a biotech firm in Cambridge, Massachusetts. Finding the corporate world "quite restrictive," Deng in 2001 joined Beijing University, where he is trying to coax human embryonic stem cells to differentiate into beta cells for treating diabetes. "Stem cell biology is a new field, so China is at the same starting line as everybody else," says Deng.

Some scientists try to keep a foot in both worlds, before deciding that China is where they want to be. In 1997, Jin set up a field station at Fudan, his alma mater, to collect DNA from China's diverse populations. In 2003, he was made dean and began splitting his time between continents. "It was really stressful to maintain two laboratories," he says. In 2005, Jin resigned from Cincinnati and moved with his family to Shanghai.

There are downsides for midcareer returnees. Salaries are smaller, for example, although a low cost of living can compensate. And whereas Chinese universities grant tenure to all faculty members, at many CAS institutes new researchers must pass reviews after several years before getting a permanent job, even if they gave up a tenured position in the West.

Middle-aged scientists also typically have families to consider. Xu recalls that his elder son had a tough time adjusting to fifth grade when the family returned from the United States in 1997. Xu's colleague, geneticist Ji Zhang, who gave up a tenured job at the University of Nebraska Medical Center in Omaha in 2002, left his wife and 17-year-old son in the States so the boy could continue his education there. "It would be difficult for my son now to adapt to life in China," Zhang says.

Lifestyle issues cut both ways. Jin says, half-jokingly, that he returned for the food. The best Chinese restaurants in Cincinnati can't match Fudan's student cafeteria, he notes. On the other hand, he and his family squeezed into an apartment one-tenth the size of their Cincinnati home. "I've been trying to convince my kids that it's not quite right for just a few people to have lived in such a big house," he says. For Jin and other midcareer returnees, cramped apartments are a small price to pay for big opportunities in China's growing research enterprise. **–DENNIS NORMILE**

Poo," says He. "He is really dedicated, working hard, and doing a good job."

But critics maintain that part-timers such as Poo are rare; many appear to leverage their own projects by taking advantage of China's abundant student labor. In the late 1990s, Xingwang Deng, a molecular biologist at Yale University, proposed using Beida's "human resources" to search for all the genes of the model plant *Arabidopsis*. The idea appealed to Gu Xiaocheng, a senior biologist, and Chen Zhangliang, then a Beida vice president; the university provided lab space and seed funds. At Yale, Deng taught a young Beida scientist, Qu Li-jia, how to make *Arabidopsis* mutants.

For his efforts, Deng was appointed a 9-month Changjiang professor by Beida, although he made clear he could not work full-time in China. To reconcile his commitments to Yale and Beida, Deng came up with a "win-win solution," says Gu: Deng persuaded Yale and Beida to establish the Peking-Yale Joint Center for Plant Molecular Genetics and Agro-biotechnology. Under Deng's directorship, the center has been generating data for the *Arabidopsis* Mutants Database and papers, most of which list Deng as senior author.

Given China's "low level" of science, Gu says, this kind of arrangement can be beneficial. "You may call it outsourcing," she says, but the resulting exchanges might not have happened otherwise. Qu adds that before the Peking-Yale Center was set up, "we did not even know how to grow *Arabidopsis*, but now seven labs at Beida do related work."

Other part-timers are following Deng's example. In 2002, Tian Xu, a Howard Hughes Investigator at Yale and a Changjiang *jiangzuo* at Fudan University in Shanghai, created the Fudan-Yale Biomedical Research Center, which now employs 20 grad students, one postdoc, and more than 40 staff to screen for genes in fruit flies and mice. And UCLA's Shuo Lin, a Changjiang *jiangzuo* at Beida since 2004, has retained a dozen grad students there to trawl for zebrafish genes.

CREDIT: CONAN LEUNG

Provocateur. Tempers flared after Harvard's Shing-Tung Yau asserted that the majority of Beijing University's overseas recruits are "fakes."

With these successes, China seems unlikely to wean itself of its part-timer dependence anytime soon. CAS is even spawning a new breed: "innovation teams" including five or six senior academics from abroad who will take turns spending a year in China and share a pot of $750,000 for research.

But Yau and other critics insist that the popularity of these programs does not justify the expense. Rather than lavish money on part-time academics, they argue, Chinese institutions should raise stipends of students and young researchers from their present paltry levels of $30 to $160 a month. "The Chinese government does not pay enough attention to young people," Yau says. As long as the brightest young minds seek greener pastures outside China, the brain drain—and the hunger for overseas talents—will continue. **–HAO XIN**

With reporting by Dennis Normile.

ORNITHOLOGY

The Pink Death: Die-Offs of the Lesser Flamingo Raise Concern

Researchers are investigating whether mass bird deaths are linked to environmental changes in East Africa's lakes

NAIROBI, KENYA—An aviator once described Lake Nakuru as "a crucible of pink and crimson fire," with a million flamingos painting an astonishing band of color that burst into pieces as the birds took flight. Such breathtaking scenes still exist in East Africa, but large numbers of its Lesser Flamingos have been dying mysteriously at Nakuru and other naturally alkaline Rift Valley lakes where they feed.

In recent months, more than 30,000 of the birds have been found dead at Nakuru, leaving enough pink carcasses to spur a newspaper to describe the lake as a "flamingo death camp." Two years ago, 43,800 of the birds perished at Tanzania's Lake Manyara, the first major die-off at that alkaline, soda-rich lake. Previous mass die-offs occurred at Lake Nakuru and two other Kenyan lakes in 1993, 1995, and 1997, as well as at two lakes in Tanzania in 2002. At the same time, birds have been gathering in places they have never been seen before. This month, thousands of Lesser Flamingos suddenly appeared at small Lake Oloiden in Kenya for the first time.

As scientists investigate what is behind the deaths and shifts in feeding sites, conservationists are worried about possible new threats from human activity that could degrade the birds' primary breeding site: Tanzania's otherworldly Lake Natron. The remote halophytic lake—whose hot, caustic waters provide a perfect nesting sight for flamingos and protect them from predators—has been proposed as site of a new baking-soda plant whose pipeline "could have quite a disastrous effect on [Natron's] water levels, which are critical for successful breeding," warns ornithologist Neil Baker, who heads the Tanzania Bird Atlas project.

Next week, two dozen of the world's leading flamingo experts will gather at a workshop in Nairobi to draft an action plan to protect the Lesser Flamingo species (*Phoeniconaias minor*). They plan to submit their recommendations to the member governments of the African-Eurasian Migratory Waterbird Agreement and the Convention on Migratory Species, which—if approved—could lead governments to take new measures.

Occasional flamingo die-offs occur naturally and are not necessarily alarming. But ornithologist Brooks Childress, who will chair the meeting, warns: "What is worrying is that the frequency of the [die-off] events appears to have increased markedly in recent years, even in relatively pristine lakes. We need to find out what the cause might be."

Suspect toxins and pathogens

Snapping on rubber gloves, German phycologist Lothar Krienitz sloshes across the muddy shore of Lake Oloiden toward a clump of pink feathers. As his hip boots sink into the muck, he reaches out a pole and hooks the flamingo carcass for later dissection and tissue sampling. Then he moves slowly to the water's edge and takes samples of the lake water with its abundant greenish growth.

Krienitz, a research associate with the Leibniz Institute of Freshwater Ecology in Berlin who collaborates with researchers at Kenyatta University in Nairobi, has been sampling the alkaline waters of the Kenyan flamingo lakes since 2001 and looking for changes in salinity, cyanobacterial species, toxins, and other factors that might explain flamingo die-offs. His institute also analyzes the flamingo tissue samples for evidence of fatal toxicity.

"There are strong indications that cyanobacterial toxins are contributing to the mass flamingo deaths," Krienitz says, cautioning that the poisonous substances, including the neurotoxin anatoxin-a as well as several liver toxins, may be only one of several explanations for the die-offs. He and others argue that shrinking lake levels and changing salinity have led to a different mix of cyanobacteria, some of which produce toxins that can kill the pink birds.

Inyasi Lejora of Tanzania National Parks says that scientists at the University of Dar es Salaam's aquatic sciences faculty are convinced that the massive flamingo die-offs at Lake Manyara in 2004 were caused by "cyanobacterial toxins." But Lake Nakuru's senior research scientist, Apollo Kariuki, says an analysis of bird tissues by Kenya's National Veterinary Laboratory implicates a pathogenic bacterium, *Pseudomonas aeruginosa*, in the current die-off. Other experts questioned that conclusion and called for further studies of algal toxins.

Lesser Flamingos, the smallest but most numerous of the world's six flamingo species, are notoriously selective in their feeding habits. They fly from one Rift Valley soda lake to another to find abundant sources of their primary food: a cyanobacterium called *Arthrospira fusiformis*, which the birds scoop into their filtering bills.

Ecologist David Harper of the University of Leicester, U.K., who leads the Earthwatch Institute's Lakes of the Rift Valley project, believes the emergence of cyanotoxins as well as the apparent increased vulnerability of the flamingos to disease are both linked to environmental changes in Rift Valley lakes. Water diversion has lowered several lakes' volumes,

CREDIT (TOP): TIM DAVIS/GETTY IMAGES

while pollutants and a host of alien flora and fauna have changed their ecology.

Other theories about the causes of the flamingo die-offs abound; various scientists have attributed the mass deaths to avian tuberculosis or cholera, botulism, heavy-metal pollution, pesticide runoffs, or combinations of such factors. Childress—global coordinator of the Flamingo Specialist Group, associated with both the World Conservation Union's Species Survival Commission and Wetlands International—suspects that toxins are responsible for killing the birds, perhaps by making them more susceptible to infectious diseases.

Veterinary microbiologist Lindsay Oaks of Washington State University, Pullman, who helped solve the mystery of *Gyps* vulture die-offs in India and Pakistan 2 years ago (*Science*, 16 June 2006, p. 1591), says "systematic and more comprehensive investigations" are needed to determine what is killing the flamingos. Val Beasley, a veterinary toxicologist at the University of Illinois, Urbana-Champaign, who helped organize a 2004 symposium on flamingo die-offs, agrees that more research is needed "to determine if algal toxins, metals, or other stressors weaken the birds so that infections become more widespread and more lethal."

Hot water

The highly alkaline Lake Natron, whose waters can reach temperatures as high as 41°C, is so remote that it was not until 1954 that biologist Leslie Brown discovered that it was the only regular breeding ground for Lesser Flamingos in East Africa. That remoteness, as well as the fact that few predators brave the hot, caustic waters to attack the chicks, may be a primary reason why the flamingos flock to Natron to breed on islands of dried mud. The birds breed infrequently—some estimate once every 5 years—and when chicks mature, they fly off to feed in other lakes.

Conservationists worry that Natron, which is not in a national park, could be damaged if two development projects succeed: a proposed large plant that would harvest and pipe away the valuable soda, and separate plans to build several tarred roads, making the area more accessible to hunters and tourists.

The proposed soda plant at Lake Natron would likely be a modern version of the current plant at nearby Lake Magadi in southern Kenya. In 1962—in what may have been the only major breeding event of East African Lesser Flamingos outside of Lake Natron in the last half-century—some of the birds for unknown reasons tried to breed in Lake Magadi with disastrous results. The lake water levels receded so quickly because of drought that soda deposits congealed on the legs of the vulnerable chicks, killing many; others were saved by a rescue effort.

Jasson John of the Wildlife Conservation Society of Tanzania, a nongovernmental organization, does not draw parallels with Lake Magadi but worries that "a large soda plant at Lake Natron might interfere with the water levels and other ecological conditions of the lake." John was among the 20 Tanzanian flamingo experts, wildlife officials, tourist industry representatives, and scientists who gathered last month in Dar es Salaam to evaluate the nation's flamingo population and discuss conservation plans that will be presented at next week's meeting of international flamingo specialists in Nairobi.

One big challenge for them is to agree on the population of Lesser Flamingos living in East Africa. Estimates have ranged from about 1.5 million to 4 million, but the census is complicated by the birds' night movements between alkaline lakes as well as the fact that Tanzania counts it birds at a different time than does Kenya. The species is classified as near-threatened because it is dependent on a very small number of unprotected breeding sites and has stringent limitations on its breeding conditions.

Childress, who has tracked several Lesser Flamingos using satellite transmitters, says scientists do not know exactly why the birds move so often from one location to another, but the search for food is not the only explanation. "The tagged birds often left a location with plenty of cyanobacteria, leaving hundreds of thousands of other Lesser Flamingos still at the location and still feeding normally," he says.

Lately, it seems that some of the flamingos have been behaving almost like tourists, visiting new places along the Rift Valley. Earlier this month, Harper stood on the shore of Lake Oloiden and marveled at the sudden arrival of several thousand Lesser Flamingos—the first time in his dozen years of studying in the region that the birds have appeared there. Krienitz's measurements this month confirmed that the shrinking of the lake has led to higher salinity, which in turn has nourished cyanobacteria, attracting flamingos.

Although East Africa has by far the largest population of Lesser Flamingos, there are also smaller colonies in West Africa and southern Africa. In Botswana, ornithologist Graham McCulloch lists disturbance from tourists—including low-flying aircraft—among the potential threats to the breeding of the colony of about 150,000 Lesser Flamingos in the Sua Pan area. But the birds' status as an undeniable tourist draw also may work in their favor. Even if governments are slow to respond to environmental or scientific arguments, concerns about tourism, a leading industry in Kenya, Tanzania, and Botswana, may compel actions needed to preserve and protect the flamingo. Before any such measures can be taken, however, scientists must first solve the mystery of what is killing these exotic birds, whose feathers have brought astonishing hues to the Rift Valley Lakes for millennia. **–ROBERT KOENIG**

Sleuth. Knee-deep in mud, Lothar Krienitz collects a dead flamingo for tissue sampling at one of Kenya's Rift Valley lakes.

CREDIT: DORIS KRIENITZ

MANTLE DYNAMICS

Rising Plumes in Earth's Mantle: Phantom or Real?

Seismologists probing the planet's depths are generating tantalizing images, but whereas some researchers see signs of plumes feeding volcanic hot spots, others see noise

Almost half a century after the plate tectonics revolution, geoscientists still have a hangover. By 1970, a decade or two of geophysical observation beneath the world's oceans had ushered in jostling plates and sinking slabs. But plate tectonics can't explain midplate volcanic centers such as Hawaii, and a hypothesis proposed in 1972—that hot rock rises in narrow plumes through the mantle to stoke such hot spots—still gets a mixed reception. "The existence of plumes is controversial to some and old hat to others," geophysicist Norman Sleep of Stanford University in Palo Alto, California, recently noted. "Skeptics are justified in demanding deep evidence for a deep-mantle hypothesis."

The latest evidence from the deep—pictures of the interior painted with seismic waves—is stirring up a field in which such tomographic results have often been disputed. At the same time, geological and geochemical studies are bringing some putative plumes into question (*Science*, 8 September, pp. 1394 and p. 1426). The new imaging will not calm the turmoil anytime soon, but it is forcing seismic tomography researchers to grapple with the limitations of their tools.

The hubbub in global seismic imaging started when a group of Princeton University seismologists introduced a new analytical tool to sharpen their view. The problem with plumes has been that according to theory they would be narrow, perhaps a couple of hundred kilometers across at most. A hot plume would slow the part of a seismic wave that passes through it from an earthquake to a seismometer. But the slowed segment of the wave—which in a tomographic analysis would paint a splotch of warm mantle in the image—could then "heal" before ever being recorded, much as an ocean wave can reform after passing around the piling of a pier.

So Raffaella Montelli, then a graduate student at Princeton and now at ExxonMobil in Houston, Texas; her Princeton adviser Guust Nolet; and theoretical seismologist Anthony Dahlen of Princeton developed a way of analyzing seismic data that for the first time takes account of such wave behavior. In their version of "finite-frequency" analysis, Montelli and colleagues were able to combine so-called P (for primary) seismic waves of two frequencies to form an image of the global mantle from a high-quality data set. Where others had reported nothing more than a debatable plume or two beneath Hawaii and Iceland, the Princeton group saw plumes of varying height beneath most of the classic hot spots, 32 plumes in all (*Science*, 5 December 2003, p. 1643).

With the proliferation of plumes and the introduction of a radically new technique, the plume debate only intensified, so Montelli and colleagues have gone one step further. In a paper in press in *Geochemistry Geophysics Geosystems*, they report how they formed a new global image from S (secondary) waves rather than P waves, again using their finite-frequency technique. S waves—which have a shearing or twisting action—react differently to variations in rock temperature and composition than do P waves, which are compressional, like sound waves. But almost all of the plumes they saw in the P-wave image they also found in the S-wave model. "There is remarkable agreement," says Nolet.

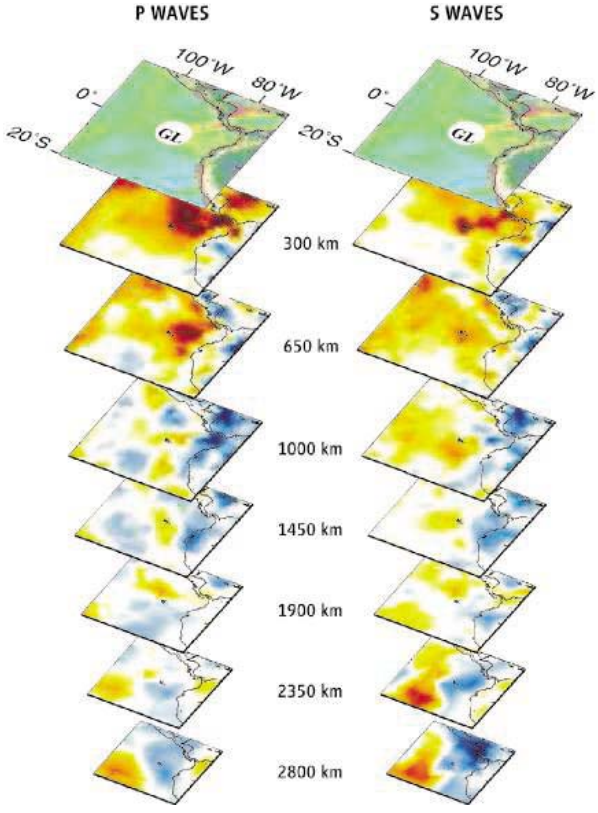
The geophysics community's reaction has been mixed. "I must say I found it striking that with both S and P they do get very similar images for some of the plumes," says theoretical seismologist Jeroen Tromp of the California Institute of Technology in Pasadena. To some others, the picture is much fuzzier. "There are similarities, but many differences too," says seismic tomographer Rob van der Hilst of the Massachusetts Institute of Technology in Cambridge. And tomographer Adam Dziewonski of Harvard University simply says that "it's difficult to argue these things are real" in either P or S renditions.

A fundamental problem, say many researchers, is a dearth of data. Everyone agrees that some finite-frequency technique is the way to go, but many argue that even it is being overwhelmed by the limited data available. Most earthquakes that seismically light up Earth's interior fall around the Pacific's Ring of Fire, whereas the seismometers recording them are limited to the continents and a few islands. So even data sets drawing on millions of quake recordings leave parts of the mantle largely in the seismic dark. As a result, "there are infinitely many [tomographic] pictures of Earth that all satisfy the data," notes Nolet.

To sort out which picture is the most likely one, the analyst must twist some knobs on the procedure to sharpen the picture while keeping things physically realistic. "This is sort of like reading tea leaves," notes Dziewonski. And methods for quantitatively gauging how well the final picture can explain the data are still severely limited by computer power. "Interpretation of tomographic models [of the mantle] is a high-risk operation," concludes Dziewonski.

To reduce the risk, researchers, predictably, call for more and better data. New seismometers are filling gaps in coverage, but it's taking longer to incorporate new kinds of data from existing seismic records. Most observers see another 5 to 10 years before they'll be able to say with confidence whether plumes exist after all.

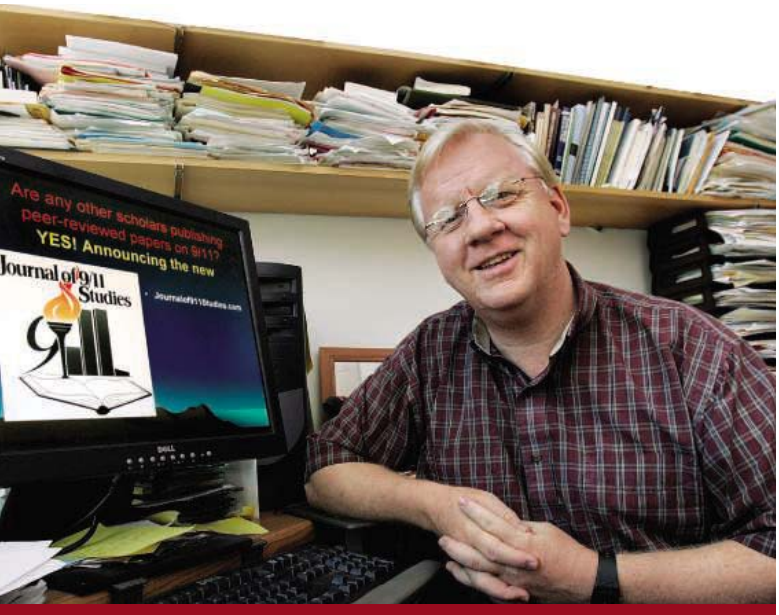
–RICHARD A. KERR



A deep plume? Extra-hot rock (red and yellows) appears as deep as 2000 kilometers beneath the Galápagos Islands in images generated from either P seismic waves or S seismic waves.

CREDIT: R. MONTELLI *ET AL.*, *GEOCHEMISTRY GEOPHYSICS GEOSYSTEMS* (2006 BY THE AMERICAN GEOPHYSICAL UNION)

EDITED BY YUDHIJIT BHATTACHARJEE



ON CAMPUS

MINORITY REPORT. A physics professor at Brigham Young University (BYU) in Provo, Utah, was placed on paid leave this month in connection with controversial statements and writings he has made on the 2001 destruction of the World Trade Center in New York City. Steven E. Jones is among a small group of scientists who cite photos, material evidence, and lab experiments to advance the hypothesis that explosive devices planted inside the towers—perhaps by the U.S. government—are what caused their destruction.

Jones's work on the subject includes a recent paper in the online *Journal of 9/11 Studies*, which he co-edits. That paper includes a disclaimer labeling it "the sole responsibility of the author." But the university is anxious to dissociate itself from Jones's hypothesis, saying it has "not been published in appropriate scientific venues."

The school is looking into whether Jones has sufficiently clarified when he is speaking for himself and not the university. Eric Combest of the Washington, D.C.–based American Association of University Professors says BYU's actions are a "fairly egregious violation of academic freedom." Jones declined to comment.

<< They Said It

"As administrator, I put the Hubble Servicing Mission back into our science plan. I rebalanced the science portfolio out of respect for National Academy priorities and out of concern for the health of important disciplines like earth science and heliospherics. ... So what's all the tumult and shouting about? A few key things come to mind: ... money, respect, and power."

—NASA Administrator Michael Griffin in a 12 September talk followed by a discussion at Goddard Space Flight Center in Greenbelt, Maryland. He said his remarks were intended to "reduce some of the angst in the [scientific] community" about his recent controversial moves.

NONPROFIT WORLD

NURTURING TALENT. The new president of the Academy of Sciences for the Developing World says stemming brain drain from developing countries will be one of his top priorities. The 700-member academy is headquartered in Trieste, Italy.

Jacob Palis, a 66-year-old Brazilian mathematician who was elected to the post this month, promises that he will work hard during his 3-year term to get scientifically advanced nations to help improve graduate and postdoctoral education elsewhere. "We must also take advantage of the growing scientific proficiency of such developing countries as China, India, and Mexico," he says, to build the capacities of the world's poorest countries.

Another priority for Palis, a researcher at the Institute of Pure and Applied Mathematics in Rio de Janeiro, will be increasing the participation of women in science, especially in leadership positions. Palis succeeds Indian materials scientist C. N. R. Rao, who completes his term in January.

NAE PRESIDENT. Former Massachusetts Institute of Technology president Charles Vest has been nominated to lead the National Academy of Engineering. A mechanical engineer with a long career in administration and policy, Vest, 65, serves on the President's Council of Advisors on Science and Technology. He says he wants to "inspire a new generation of young men and women to pursue careers in engineering to improve the quality of human life and strengthen the American and worldwide economies." If elected—he is the unanimously chosen nominee for the position—Vest will begin a 6-year term in July 2007, succeeding William Wulf.

IN THE COURTS

VIOLENT ACTIVISM. Three animal-rights activists have been handed prison sentences for terrorizing U.S.–based employees of a British life sciences company. The trial, heard before a federal court in Trenton, New Jersey, was the first to be conducted under a new antiterrorism provision that was added to the Animal Enterprise Protection Act in 2002. The activists are members of Stop Huntingdon Animal Cruelty (SHAC), which has been running a global campaign against Huntingdon Life Sciences. The company tests drugs on animals at centers in the United Kingdom and in Princeton, New Jersey.

In a 12 September ruling, U.S. District Judge Anne Thompson found the three guilty of stalking Huntingdon employees and of provoking threats and vandalism against them. She sentenced Kevin Kjonaas, 28, to 6 years' imprisonment, Lauren Gazzola, 27, to 4 years and 4 months, and Jacob Conroy, 30, to 4 years and ordered them to pay $1 million in damages to the company.

Got a tip for this page? E-mail people@aaas.org

CREDIT: PAT OLMERT/NSF

JUDGES (left to right)

Felice Frankel
Research Scientist
Massachusetts Institute of Technology
Cambridge, Massachusetts

Corinne Sandone
Medical Illustrator, Assistant Professor
Art as Applied to Medicine
Johns Hopkins School of Medicine
Baltimore, Maryland

Donna Cox
Director, Visualization and Experimental Technologies
National Center for Supercomputing Applications
Urbana, Illinois

Michael Keegan
Assistant Managing Editor, News Art
The Washington Post
Washington, D.C.

Thomas Lucas
Thomas Lucas Productions Inc.
Ossining, New York

2006 Visualization Challenge

The still life on the cover of this week's issue of *Science* is not a photograph but a computer-generated rendering of five famous mathematical surfaces. The result, created by Richard Palais of the University of California, Irvine, and graphic artist Luc Benard, is a virtuoso display of modern computer-graphics technology. (Notice how the glassy surfaces are reflected in one another and in the glass-covered, wood-grained tabletop.)

The image is the first-place winner in the illustration category of the 2006 Science and Engineering Visualization Challenge. Indeed, it is a prime example of why *Science* and the National Science Foundation (NSF) have jointly sponsored the visualization challenge every year since 2003. It is beautiful. It can capture the imagination of nonscientists. But it also represents a powerful new tool for research. As Benard and Palais wrote in their application, "Mathematicians have always needed to 'see' the complex concepts they work with in order to reason with them effectively. In the past, they conjured up mental images as best they could, but the wonders of computer graphics provide them with far more detailed pictures to think with." Or, as Felice Frankel of the Massachusetts Institute of Technology, one of the challenge judges, put it, "The science community needs to discuss the enormous contribution good visual translations can bring to both communication and advancing the thinking behind the science. Critically thinking about what makes an honest and successful representation and raising our standards can only be beneficial for the science community as a whole."

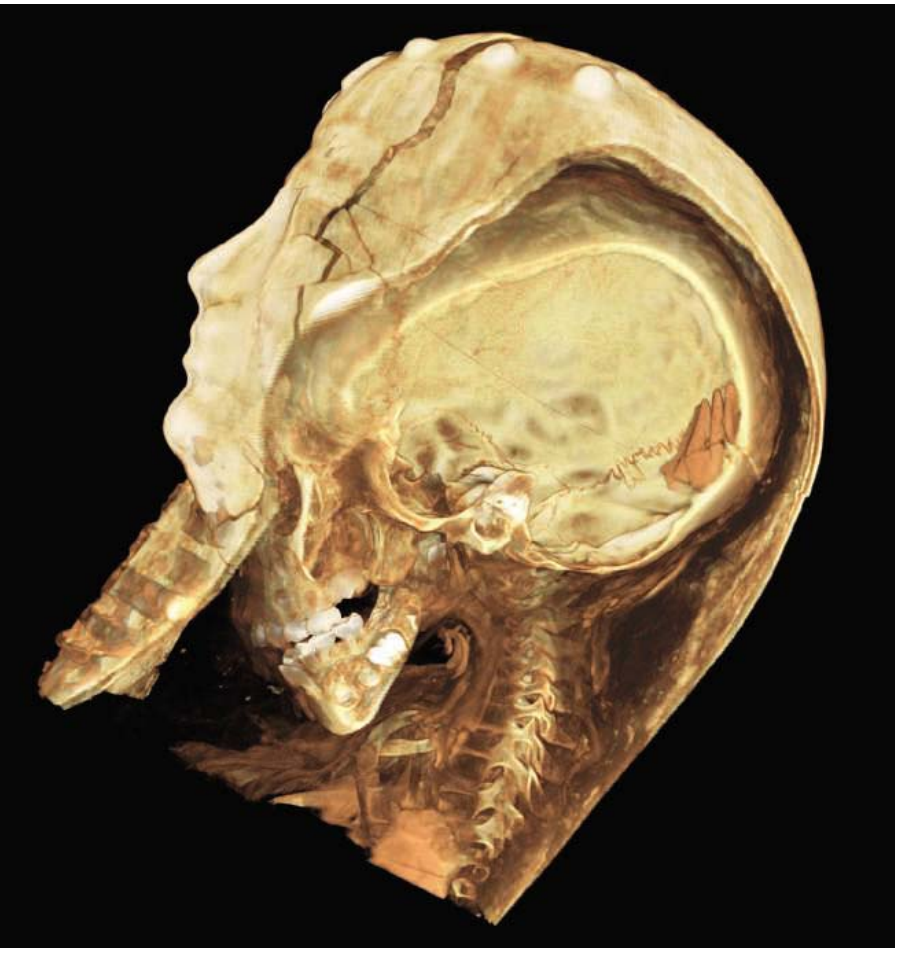
The visualization challenge is intended to showcase and encourage this kind of work. This year, we invited submissions in five categories: photography, illustration, informational graphics, noninteractive multimedia, and interactive multimedia. Entries were screened by a committee from NSF and *Science*. Then an independent panel of experts in scientific visualization reviewed the finalists and selected the best, which appear in these pages.

We urge you and your colleagues to contribute to the next competition, details of which will be available at www.nsf.gov/news/special_reports/scivis/index.jsp, and to join us in congratulating the winners and all the other entrants.

Susan Mason of NSF organized this year's challenge, Rhitu Chatterjee of *Science*'s news staff wrote the text accompanying the images in the following pages, and *Science*'s online editors Stewart Wills and Tara Marathe put together a special Web presentation at www.sciencemag.org/sciext/vis2006.

Jeff Nesbit, Director, Office of Legislative and Public Affairs, NSF
Monica Bradford, Executive Editor, *Science*

2006 Visualization Challenge



FIRST PLACE

An Egyptian Child Mummy

Robert Cheng, W. Paul Brown, and Rebecca Fahrig, Stanford University, and Christof Reinhart, Volume Graphics

For 75 years, this child mummy resided in the Rosicrucian Egyptian Museum in San Jose, California, its body unseen by human eyes, its story a mystery. Then, in early 2005, a team of researchers and computer engineers led by W. Paul Brown of Stanford University began to unravel the threads of this mystery using the latest imaging technology.

Radiologists at Stanford University used a high-resolution C-arm computed tomography (CT) scanner from Siemens Medical Solutions to generate 60,000 2D scans of the unopened, intact mummy. Computers running the latest 3D computer graphics at Silicon Graphics used these scans to create a 3D model of the mummy and its interior.

Analysis of the data revealed that the 2000-year-old mummy is the remains of a 4- or 5-year-old girl from a well-to-do family. The Rosicrucian museum has since named her Sherit, ancient Egyptian for "Little One." Her body showed no telltale signs of trauma or long-term disease, and so the researchers believe Sherit died unexpectedly. "She must have died from an infectious disease," says Brown, nicknamed "Mummy Daddy" by his team members. "We want to put together a CD to send it to different museums."

Felice Frankel, a member of the panel of judges, says that the panel's decision was undeterred by the fact that CT scanning and computer imaging, rather than traditional photography, produced the "stunningly beautiful" image. It shows how "the definition of photography in science has expanded," she says.

Photography

SECOND PLACE

Cockroach Portrait

David Yager, University of Maryland

Cockroach haters, look your enemy in the eye! Photographing small animals like this 2-centimeter-long Cuban banana cockroach, *Panchlora nivea*, has its challenges: You can focus on only a small part of the tiny animal in one shot. To overcome this drawback, David Yager of the University of Maryland, College Park, relied on technologies old and new. He laid the dead roach on its back on a bed of glass beads and took multiple snapshots at different depths of field through a regular dissecting microscope. Each frame focused on different parts of the roach's head. With three light tubes, he lit the roach's face from various angles and peeked into its head. Next, he merged 12 separate frames using image-processing software called Automontage to create a clear and detailed "Cockroach Portrait."

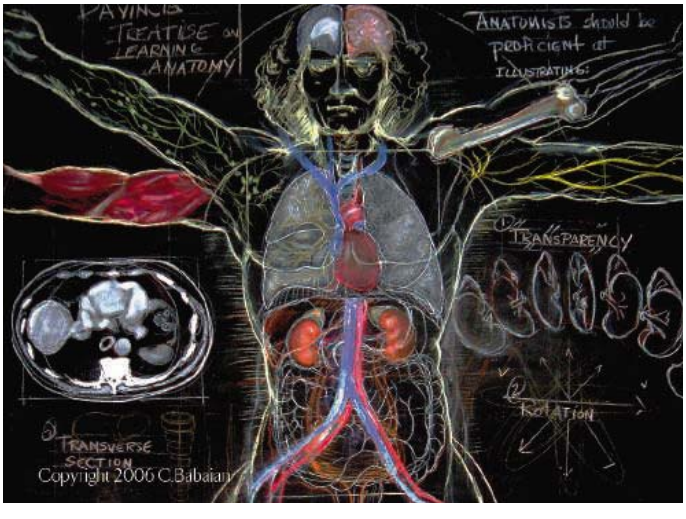
Illustration

SECOND PLACE

A Da Vinci Blackboard Lesson in Multi-Conceptual Anatomy

Caryn Babaian, Bucks County Community College, Newtown, Pennsylvania

Some things never grow old. Leonardo da Vinci's *Vitruvian Man*, first drawn more than 500 years ago, is still teaching people about the intricacies of the human body. Biology teacher Caryn Babaian of Bucks County Community College in Newtown, Pennsylvania, uses the iconic sketch as a "multi-conceptual image" in her introductory anatomy class to illustrate three crucial anatomical concepts: rotation, transparency, and transverse section. Babaian requires her students to draw the image in their notebooks as they watch it take shape on the blackboard. Panel of judges member Thomas Lucas says even though the use of the image "gave inspiration to a few people, the effect on them might have been more powerful than something that went over the mass media."



FIRST PLACE

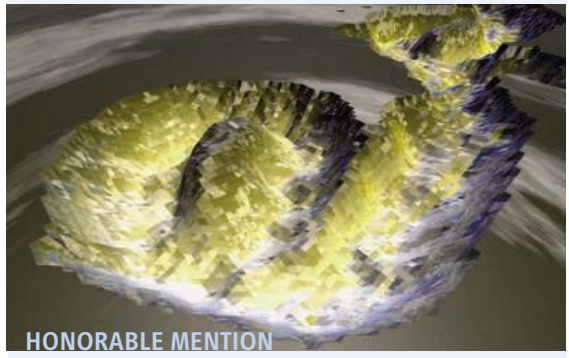
Still Life: Five Glass Surfaces on a Tabletop

Richard Palais, University of California, Irvine, and Luc Benard

To most of us, a surface is something we can touch and attribute a shape to—the spherical surface of a ball or the toroidal surface of a doughnut. But there are innumerable surfaces that we cannot touch, or see, or even know of, because they are representations of mathematical functions. Mathematicians have long relied on their own powers of imagination to picture these abstract surfaces. Now, mathematicians such as Richard Palais of the University of California, Irvine, and graphic artists such as Luc Benard are exploiting the magic of computer graphics to recreate these abstract mathematical surfaces in familiar yet intriguing settings.

This illustration presents five well-known mathematical surfaces, rendered as glass objects in a highly realistic "Still Life." To create their chosen surfaces, Benard relied on the computer program 3D-XplorMath, developed by Palais for visualizing many of the most famous mathematical surfaces. He then exported these surfaces into a 3D-rendering program, using it to give the objects a glassy texture and place them on a virtual glass-covered wooden tabletop.

Panel of judges member Felice Frankel was impressed by the image's ability to engage viewers and trigger their curiosity. "That is what we strive for in any visual experiment, that we are creating curiosity, by engaging each other [visually]," she says. "That is how we can learn from one another."

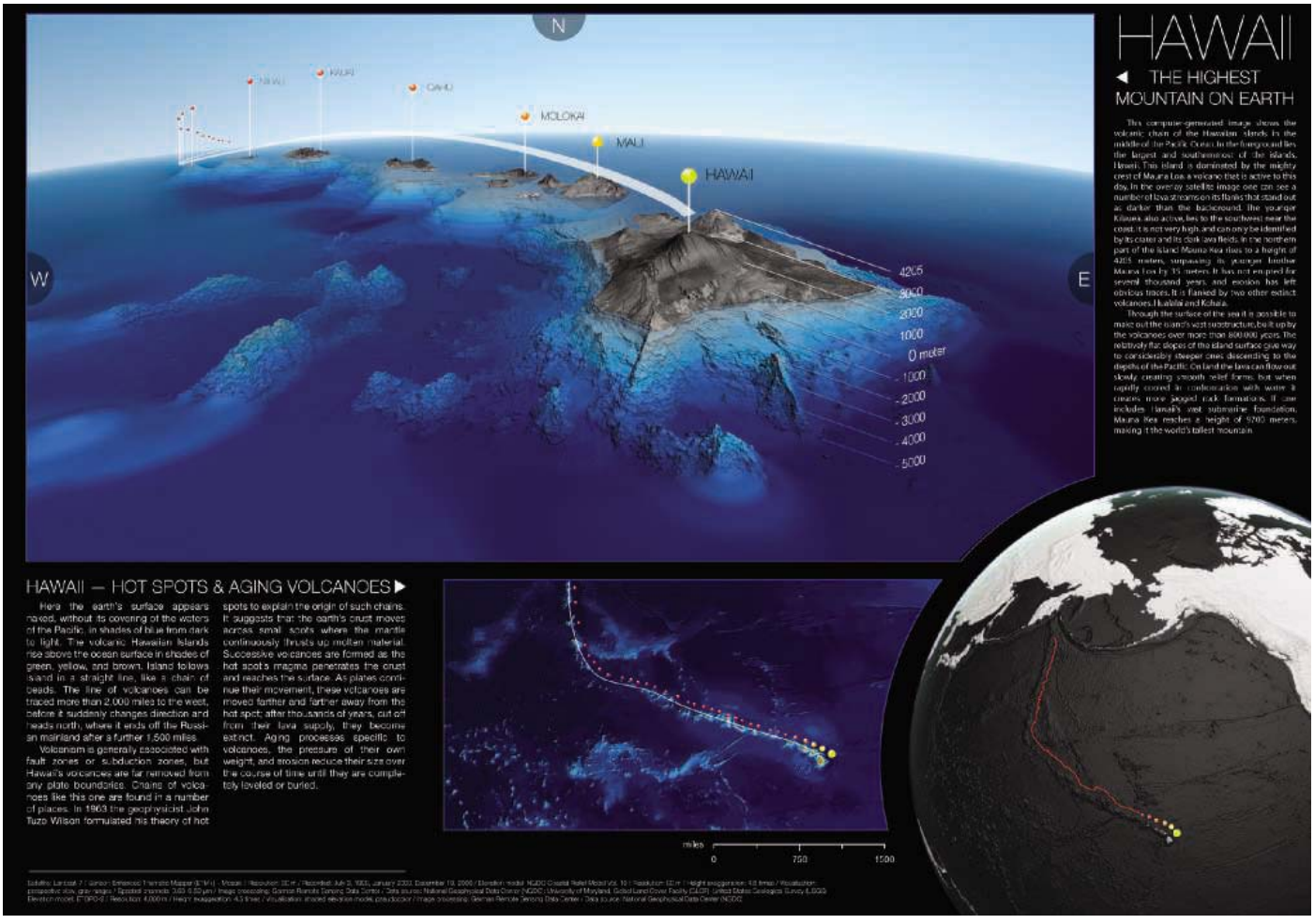


HONORABLE MENTION

The Handwritten Letter "e"

Curtis DuBois

This landscape began as an effort to aid handwriting analysts. According to independent media artist Curtis DuBois, who is based in Lummi Island, Washington, "every individual has a characteristic way of using pressure in their writing," resulting in a unique pattern of pressure points. He used a 3D ray-tracing program to convert the shades of gray in a digitally scanned image of the handwritten letter "e" into variations in virtual altitude. By turning the darker spots into deeper areas in the image, DuBois was able to highlight the "hot spots" or pressure points and thus increase the amount of information available in the writing. He then added color and "atmospheric effects" for "aesthetic impact" of the image.



FIRST PLACE

Hawaii, the Highest Mountain on Earth

Nils Sparwasser, Thorsten Andresen, Stephan Reiniger, and Robert Meisner, German Aerospace Center

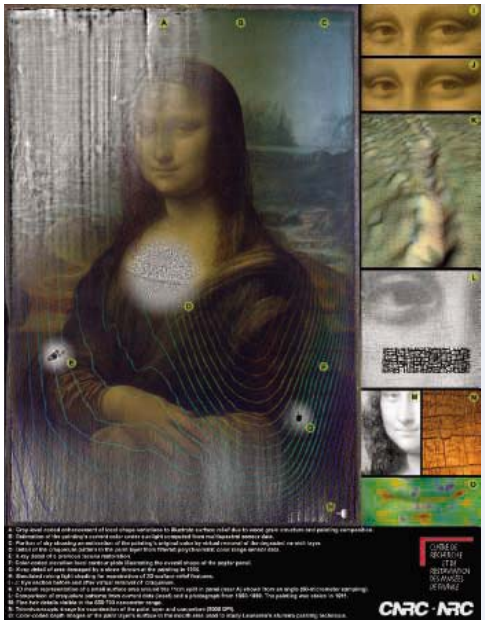
Mount Everest is the highest mountain on Earth above sea level, but it's not the world's tallest. That honor goes to the Hawaiian volcano Mauna Kea. When measured from its base on the Pacific Ocean floor, it is about 1000 meters taller than Mount Everest. Mauna Kea is part of a 5600-kilometer-long string of volcanoes stretching westward from the main Hawaiian island.

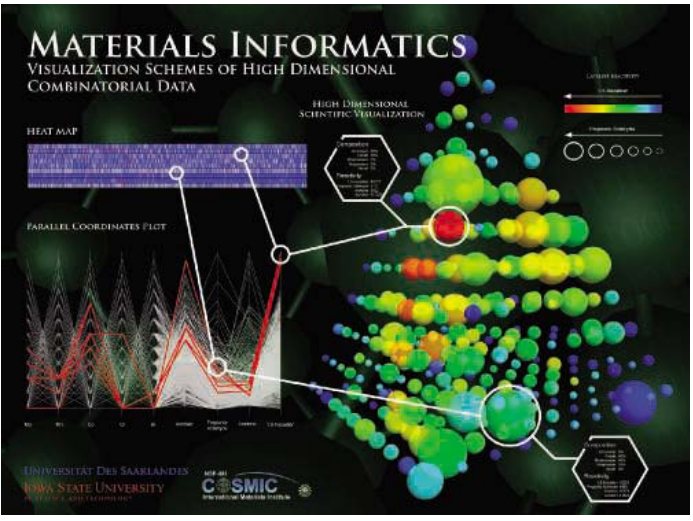
Geographer Nils Sparwasser and his colleagues at the German Aerospace Center in Oberpfaffenhofen introduce us to the Hawaiian volcanoes with this panoramic view across the Pacific Ocean. The illustration combines data gathered by satellites and ships with the latest in computer-modeling technology.

The image shows the five volcanoes on the main Hawaiian island, including Mauna Kea, with the smaller islands stretching westward behind them. The jagged submarine surfaces of the volcanoes are formed by the rapid cooling of hot lava under water.

The volcanic chain spanning the Pacific (inset) is formed by small convection streams called "hot spots," just below Earth's crust, where magma rises from the hotter parts of the mantle. These hot spots melt the tectonic plates moving above them, causing magma and bits of the molten plate to erupt onto the sea floor. Over time, the lava accumulates, forming a mountain that rises above sea level. The moving tectonic plates carry the newly formed mountain away from the original location, as newer volcanoes continue to form in the same spot.

Panel of judges member Donna Cox says she found the image "compelling" because it conveys information with ease without the viewer having "to read too much of the text."





HONORABLE MENTION

Materials Informatics

Matt Heying, Changwon Suh, and Krishna Rajan, Iowa State University, and Simone Seig, Universität de Saarland

Chemists are forever hunting for newer and more efficient catalysts. The task can require sifting through enormous amounts of data on the chemistry of potential candidates. Materials scientist Krishna Rajan and his colleagues at Iowa State University and the University of Saarland in Germany have made the job easier with this visually captivating yet comprehensible informational graphic. In a glance, a catalyst researcher can get information on the composition of thousands of catalysts and their chemical reactivity.

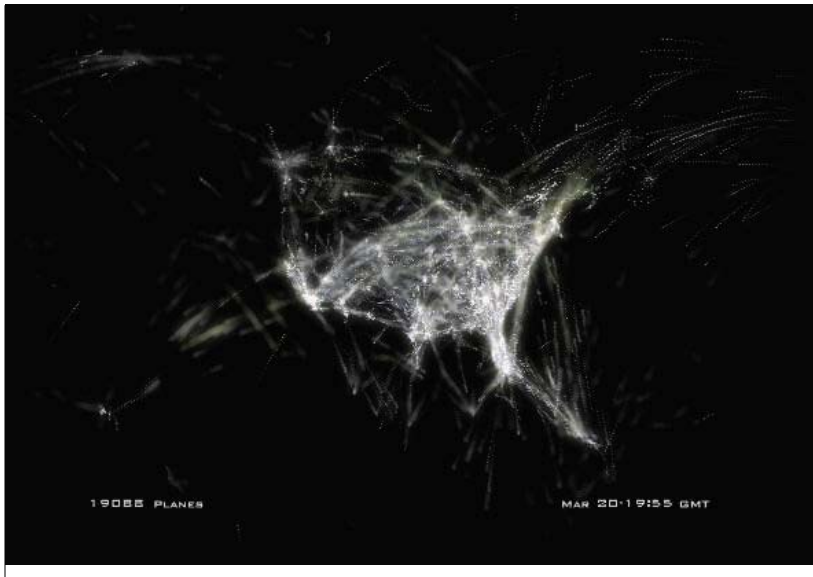
SECOND PLACE

Mona Lisa Montage

Louis Borgeat, François Blais, and John Taylor of the National Research Council of Canada, and Christian Lahanier of the Centre de Recherche et de Restauration des Museés de France

It may not be the prettiest Mona Lisa image you have seen, but it is certain to be the most informative. This "montage," jointly produced by the National Research Council of Canada and the Center for Research and Restoration of the Museums of France, depicts the information obtained by analyzing Leonardo da Vinci's painting using the latest scientific imaging technologies, such as a high-resolution 3D scanner and a polychromatic 13-band multispectral camera. Not only can such analyses help museum curators and conservation experts study the condition and authenticity of old paintings, but they also reveal the artists' techniques. This project filled in details of the unique pattern of cracks on the Mona Lisa, provided an estimation of its original color, and demonstrated da Vinci's sfumato painting technique.

2006 Visualization Challenge



Noninteractive Multimedia

FIRST PLACE (TIE)

Flight Patterns

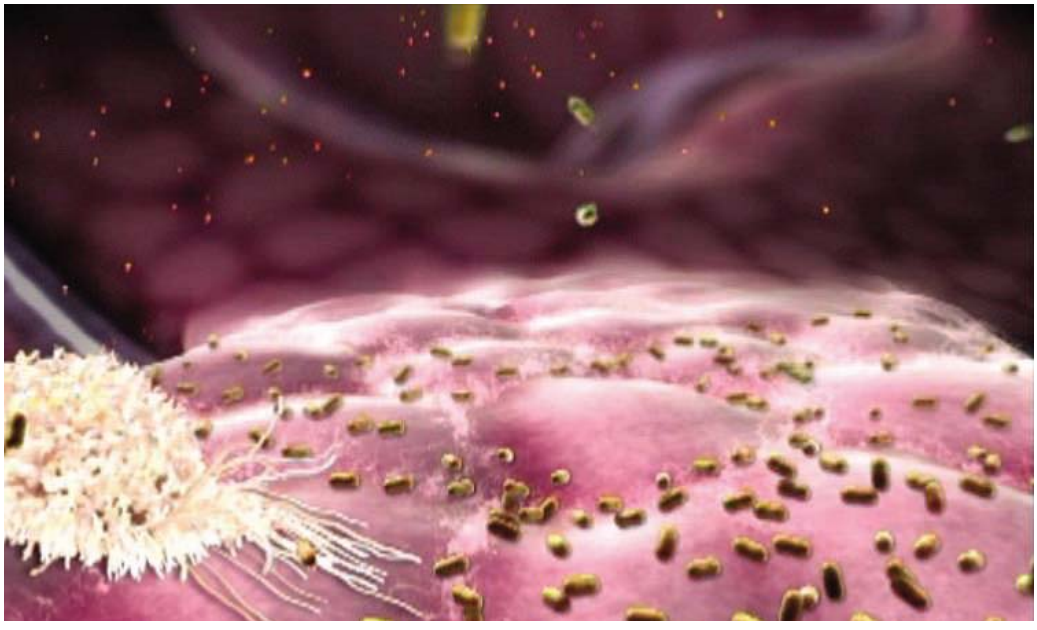
Aaron Koblin, University of California, Los Angeles

Thousands of airplanes zoom through our skies every day. Ever wondered what this air traffic looks like? In this animation made by digital media artist Aaron Koblin of the University of California, Los Angeles, it looks much like fireworks shattering the darkness of a night sky.

Using air-traffic data from the Federal Aviation Administration, Koblin shows the changing dynamics of air traffic over the United States and Canada over a 24-hour period. After categorizing the data based on criteria such as "types of aircraft," "location," and "altitude," Koblin experimented with ways to visualize the information. He plotted the data using a programming language called Processing and animated it using Adobe After Effects and Maya.

The movie begins with a splash of dotted white and thin green lines that move slowly across the screen in different directions. As the numbers and destinations of flights increase after dawn breaks across the continent, the crisscrossing lines outline the underlying landmasses of the two countries. Then the scenes repeat with more vibrant colors, eventually zooming in on New York, Los Angeles, and Hawaii.

"Not only was it incredibly informational, [it was also] unbelievably engaging," says panel of judges member Felice Frankel, who described the depiction of air traffic as "brush strokes" in a "Japanese painting." "It's one thing to convey data and another to make somebody want to look," she added.



FIRST PLACE (TIE)

Body Code

Drew Berry, The Walter and Eliza Hall Institute, Melbourne, Australia; Jeremy Pickett-Heaps, University of Melbourne; and François Tétaz

Originally created for an art gallery, this animation could easily pass for a science-fiction movie. But in reality, it is a glimpse inside our own bodies, humming with activity at every level—from molecules to cells to tissues and organs.

Made by scientific animator Drew Berry of The Walter and Eliza Hall Institute in Melbourne, Australia, time-lapse imaging expert Jeremy Pickett-Heaps of the University of Melbourne, and independent sound artist François Tétaz, this 9-minute animation takes the viewer on a wordless journey through a universe of alien structures and "factorylike" activities that keep us alive.

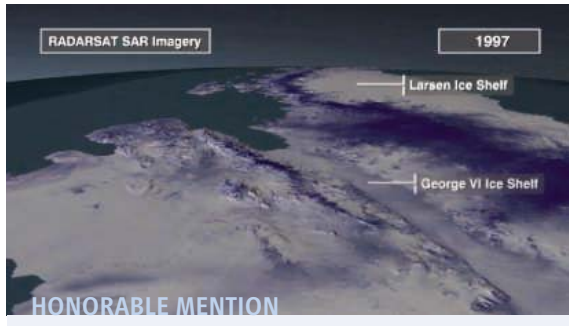
In one scene, a protein receptor sticks out of a cell's surface waiting for a messenger protein to attach itself. When that happens, the receptor transmits a message into the cell, triggering the cell to divide. Another scene takes the viewer on a roller-coaster ride through a strand of DNA with machinelike transcription enzymes, as they zip through the DNA churning out messenger RNAs along the way.

According to Berry, "time-lapse footage" had a "pivotal influence" on his reconstructions of the "living interior" of cells. He also used data from x-ray crystallography and electromagnetic tomography.

"For those of us who think cells are these static structures that divide in a passive way and pass chemicals between them passively, this was a stunning revelation," says panel of judges member Thomas Lucas, who describes the animation as a "real masterpiece."

Noninteractive Multimedia

continued ...



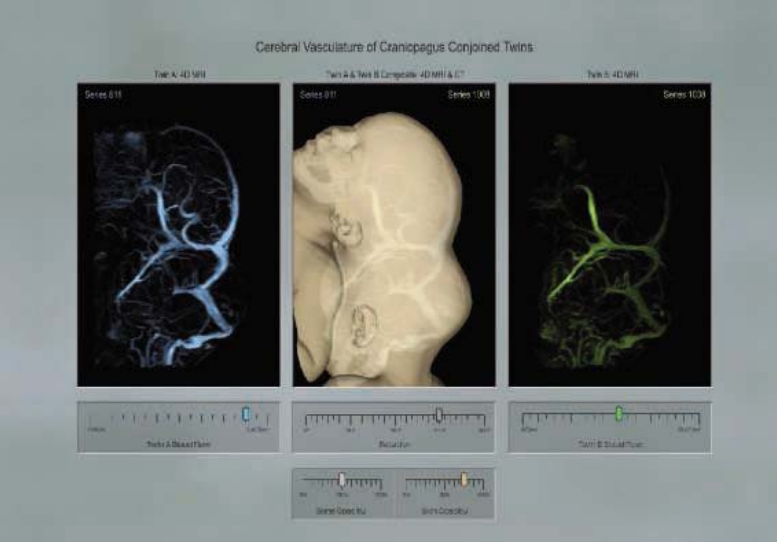
HONORABLE MENTION

A Short Tour of the Cryosphere

Jennifer Brennan, ADNET Systems Inc./NASA Goddard Space Flight Center; Waleed Abdalati and Horace Mitchell, NASA Goddard Space Flight Center; and Walter Meier, National Snow and Ice Data Center

The chain of interactions between Earth's cryosphere and its climate is endless, and this 5-minute animation gives a bird's-eye view of it all—from the crumbling Larsen B Ice Shelf in Antarctica to the shrinking sea ice in the Arctic to the seasonal ebb and flow of the snow cover in the Rockies. The movie shows the changing snow and ice cover of our planet and how these shifts could affect the global environment. Made by a group of animators and researchers at NASA's Goddard Space Flight Center and the National Snow and Ice Data Center, the animation is based on data from multiple NASA satellites.

2006 Visualization Challenge



FIRST PLACE

Cerebral Vasculature of Craniopagus Conjoined Twins

Travis Vermilye, Stephen Humphries, and Andrew Christensen, Medical Modeling, Golden, Colorado; and Kenneth Slayer, International Craniofacial Institute, Dallas, Texas

Surgeons trying to separate conjoined twins joined at their heads face a complex problem: The twins' brains are distinct, but they share major blood vessels. The challenge is to divide the blood vessels between the twins so that each has an adequate blood supply. To evaluate the chances of successfully separating one set of so-called craniopagus conjoined twins, a group of surgeons at the International Craniofacial Institute in Dallas, Texas, used an interactive tool developed by medical illustrator Travis Vermilye, medical physicist Stephen Humphries, and their team members at Medical Modeling LLC in Golden, Colorado.

The tool uses three sets of images. The ones on left and right each show the blood flow through a single twin's head. Those in the center show the twins' joint blood vessels within the framework of the twins' skeletal and facial features. The slider bars below each panel rotate the individual frames through 360 degrees—to view the images from all possible angles—and also control the opacity of the facial and skeletal tissues. Vermilye used volumetric magnetic resonance imaging scans of each twin to construct a three-dimensional view of the cranial circulation, and he relied on computed tomography scans for the skeletal and soft facial tissue animations. This tool helped the surgeons postpone the separation of the twins.

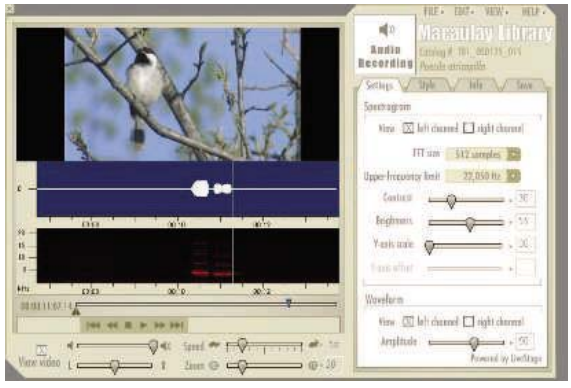
Panel of judges member Thomas Lucas says he was impressed by the "simplicity of the tool in contrast to the complexity of interactions." He adds: "It offers the layperson an incredibly fascinating glimpse into nature gone awry."

SECOND PLACE

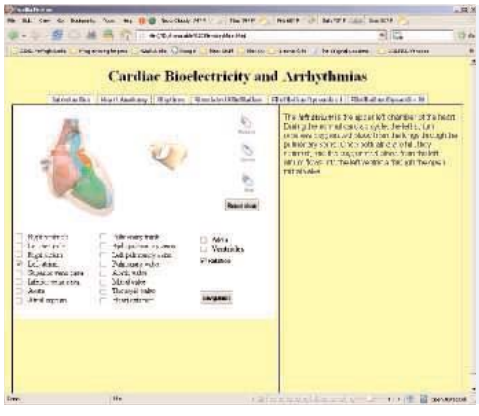
A Real-Time Audio and Video Sound Visualization Tool

Jack Bradbury, Guillaume Iacino, Erica Olsen, and Robert Grotke, Cornell Lab of Ornithology, Cornell University

For scientists and laypeople alike, this tool, made by Jack Bradbury and his team members at Cornell University, offers a unique opportunity to hear and see sounds in real time. The user can listen to sounds of all sorts of animals—including crickets, seals, whales, fish, and birds—while watching the animal on the top panel (in most cases, videos are available) and view dynamically generated waveforms and spectrograms of the sound in the middle and bottom panels, respectively. Control bars allow the viewer to change various parameters, such as the color of the spectrogram and its brightness. Intended as an educational tool, it is available free at www.animalbehaviorarchive.org and can be used by anyone with a computer, an Internet connection, and a QuickTime player.



Interactive Multimedia



HONORABLE MENTION

Cardiac Bioelectricity and Arrhythmias

Flavio Fenton and Elizabeth Cherry, Cornell University

Deep inside a human heart, its pacemaker sends out bursts of electrical signals that keep the heart pumping rhythmically, supplying life-giving oxygen to the body. When these electrical waves become disorganized, the heart starts beating irregularly or arrhythmically. Flavio Fenton and Elizabeth Cherry of Cornell University made this interactive program to educate people about arrhythmias. It presents detailed information on cardiac anatomy, normal cardiac electrophysiology, and different kinds of arrhythmias using a combination of words, pictures, and computer simulations and animations.

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LETTERS

edited by Etta Kavanagh

Debating the Cause of a Neurological Disorder

IN HIS ARTICLE "GUAM'S DEADLY STALKER: ON THE LOOSE WORLDWIDE?" (NEWS FOCUS, 28 JULY, p. 428), G. Miller presents an objective review of Cox's reformulated cycad hypothesis (*1*). This hypothesis suggests that ALS-PDC, a neurological disorder once common in the Chamorro people of Guam, is caused by eating fruit bats, who have a toxin, β-methylamino-L-alanine (BMAA), in their bodies from ingesting cycad seeds. However, the case is even less compelling than Miller suggests.

1) BMAA is present in Guamanian cycad seeds (*2*), but it is not very neurotoxic, as determined in primate studies. Spencer *et al.* administered "huge" doses (greater than 100 mg^{-1} kg^{-1} day for ~12 weeks), but they found no evidence of delayed or progressive neurodegeneration, two essential requirements for a toxin to fit the epidemiological data (*3*).

2) Banack and Cox report finding BMAA in the tissue of flying foxes collected on Guam, but they show no representative data (*4*). The selectivity of their assay is questionable and their mass analysis data are flawed. Determinations made on the dried skins of three museum specimens collected 50 years prior are of questionable relevance and are likely an artifact. It is dubious to assume that the BMAA is evenly distributed throughout the animal and that the highest value measured in the dried flesh can be multiplied by the average weight of a bat to yield the ingested dose.

A fruit bat eating a cycad seed. The Letter disputes the hypothesis that eating fruit bats that have a toxin from ingesting cycad seeds causes a neurological disorder.

3) Cox and colleagues say that they found BMAA in fixed (paraformaldehyde and sucrose) brain tissue collected from affected Guamanians and stored for years prior to analysis (*5*). Such storage would promote pronounced chemical change and these findings are likely an artifact. Notably, Montine *et al.* (*6*) found no evidence of BMAA in flash-frozen brain tissue obtained from Caucasians on the U.S. mainland nor Chamorros on Guam, regardless of the presence or absence of neurological disease.

4) There is little evidence that fruit bats were a major dietary component on Guam, and there are no reports of their consumption in either of the two other regions of high ALS-PDC incidence: Japan and west New Guinea.

The scientific community has been very receptive to the BMAA hypothesis; more than ever, the onus is now on its proponents to provide compelling and credible data.

MARK W. DUNCAN[1] AND ANN M. MARINI[2]

[1]Professor of Pediatrics, Medicine and Cellular and Developmental Biology, University of Colorado at Denver and Health Sciences Center, Aurora, CO 80045, USA. [2]Associate Professor of Neurology and Neuroscience, Uniformed Services University of the Health Sciences, Bethesda, MD 20814, USA.

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Top-Down Vs. Bottom-Up Effects in Kelp Forests

IN THEIR REPORT "STRONG TOP-DOWN CONtrol in southern California kelp forest ecosystems" (26 May, p. 1230), B. S. Halpern *et al.* conclude that these forests show strong top-down (consumer-driven) control and that bottom-up (resource-driven) control in such systems may often be overestimated.

These conclusions run counter to most of the extensive literature (*1*–*4*) on the ecology and natural history of kelp forests in southern California. There are numerous examples of the importance of storms and low nutrients over large spatial and temporal scales, especially during El Niños (*3*, *5*–*7*) but also from decadal climate shifts (*8*). Halpern *et al*. used a short-term data set that did not include El Niño–Southern Oscillation or decadal climate shifts. Moreover, they used satellite-derived offshore chlorophyll a concentration data as a measure of "resources" without establishing that these data were a good proxy for nutrients or primary production in nearshore kelp forests and despite evidence to the contrary [e.g., (*9*, *10*)].

The primary evidence for top-down effects was correlations interpreted by Halpern *et al.* as showing that spiny lobsters and Kellet's whelks were "significantly important species, likely due to their strong impacts on key grazers of kelp (urchins) and algae (limpets and snails)." There is indirect evidence that lobsters may affect urchins (*11*, *12*), but Kellet's whelk is primarily a scavenger (*13*) whose abundance has been negatively correlated with kelp forests (*14*). Neither animal has been shown to have "strong" impacts on their prey species in California kelp forests. Halpern *et al.* could think of no mechanism by which the two other significant species "control" algae. The diets of these fish indicate no such mechanism; the correlations likely result from habitat preferences (*15*). The lack of significant correlation between kelp and urchins is counter to their hypothesis but was not discussed. The analytical results may be generally misleading due to weak trophic links [e.g.,

many of the grazers eat other algae in addition to kelp, and commonly eat drift, not attached plants (*3*)]. Thus, neither bottom-up nor top-down effects were tested and the conclusions, therefore, are unsubstantiated.

MICHAEL S. FOSTER,[1]* MATTHEW S. EDWARDS,[2] DANIEL C. REED,[3] DAVID R. SCHIEL,[4] RICHARD C. ZIMMERMAN[5]

[1]Moss Landing Marine Laboratories, Moss Landing, CA 95039, USA. [2]Department of Biology, San Diego State University, San Diego, CA 92182, USA. [3]Marine Science Institute, University of California, Santa Barbara, CA 93106–6051, USA. [4]Marine Ecology Research Group, School of Biological Sciences, University of Canterbury, Christchurch, New Zealand. [5]Department of Ocean, Earth and Atmospheric Sciences, Old Dominion University, Norfolk, VA 23529, USA.

*To whom correspondence should be addressed. E-mail: foster@mlml.calstate.edu

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IN THEIR REPORT "STRONG TOP-DOWN CONtrol in southern California kelp forest ecosystems" (26 May, p. 1230), B. S. Halpern *et al.* conclude that top-down (predatory) effects are strong and more important than bottom-up (nutrient) effects in setting kelp forest community structure. They reach this conclusion through a statistical technique that examines mathematical associations among variables. Like any statistical technique that tests for correlations, it is unable to assign causality or deal effectively with highly correlated explanatory variables ("multicolinearity"). By including several highly intercorrelated predictor variables in their statistical model, it is essentially impossible to estimate bottom-up effects (*1*, *2*). For example, because nutrient concentrations are tightly correlated with water temperature in southern California (*3*), it is probably impossible to separate temperature from bottom-up effects. Furthermore, their exclusion of sites from the warmest and most nutrient-poor waters (*4*) limits the ability to detect bottom-up effects.

Statistical associations between predator abundance and aspects of community structure lead Halpern and colleagues to conclude that predators drive community structure, but they offer few plausible mechanisms. A more likely causal link, bottom-up effects driving kelp forest community structure (including predator abundance) (*5*–*7*), would produce identical statistical results. For example, the predatory kelp rockfish was identified as exerting significant "top-down control," but this fish is found almost exclusively with kelp because it is dependent on it, not vice versa (*7*).

Finally, the purported top-down effects are weak, explaining at most 20% of the variation in community structure. Other variables (e.g., water temperature and geographic location) explained 2 to 27 times more of the variation in community structure for all trophic levels but kelps (*4*). Modern statistical tests give us unprecedented ability to discover patterns in complex data sets, but such patterns can only be interpreted when combined with a sound understanding of the natural history of the system.

MARK A. STEELE,[1,2]* STEPHEN C. SCHROETER,[1] ROBERT C. CARPENTER,[2] DAVID J. KUSHNER[3]

[1]Marine Science Institute, University of California, Santa Barbara, CA 93106, USA. [2]Department of Biology, California State University, Northridge, CA 91330, USA. [3]Channel Islands National Park, Ventura, CA 93001, USA.

*To whom correspondence should be addressed. E-mail: steele@lifesci.ucsb.edu

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Response

FOSTER *ET AL.* AND STEELE *ET AL.* RAISE A number of concerns about our recent study. An important aspect of our analyses is that our dependent variables were species abundances, not the aggregate trophic values that are traditionally used. Our approach enhances the possibility of detecting either bottom-up or top-down patterns for individual or groups of species because assumptions about the nature (direct or indirect) of the relationships between species and trophic levels are unnecessary, and because it can detect compensatory dynamics within a trophic level that could eliminate aggregate top-down or bottom-up effects. Foster *et al.*'s concern about the species highlighted by our analyses likely arises from their expectation that direct trophic links must exist for the results to be valid. We were not assessing whether a "trophic cascade" existed, but instead evaluating the direction of control within communities. Indeed, our results may not have emerged from traditional approaches, and they highlight the potential importance of indirect effects in controlling community structure.

Both Letters express concern that we suggest cause and effect through correlations and not experiments. Despite reliance on correlational relationships, large-scale studies like ours have a long record of providing new insight through analysis over spatial and temporal scales beyond the reach and budget of experimental study. We focused on the hypothesized mechanisms that would be responsible for either bottom-up or top-down control—nutrient availability and predator abundance—and then determined the amount of variation explained by these two different groups of variables for algal and mid-trophic level abundances. The expected cause and effect are certainly implicit in our study, but will require significant resources before they can be tested experimentally.

Steele *et al.* are correct in noting that multicolinearity can create problems (*1*). However, our principal objective was to construct the best predictive model for both top-down and bottom-up variables, a situation in which "multicolinearity can be effectively ignored" [(*1*), p. 2811]. Nevertheless, we limited multicolinearity problems within each different predictor group by eliminating highly multicolinear variables, an approach (*1*) that acts to conservatively decrease significant results (top-down control, in our case). Furthermore, we reported "pure" top-down and bottom-up effects in table S2 and Fig. 3, which are the amounts of explained variation after eliminating the multicolinearities between the different predictor groups. Contrary to Steele *et al.*'s expectations, top-down and spatial or temperature variables were colinear while bottom-up and temperature variables were not (see table), such that adding multicolinearity to our results would have suggested even stronger top-down effects.

AMOUNT OF VARIATION EXPLAINED BY MULTICOLINEARITY BETWEEN GROUPS OF PREDICTOR VARIABLES

		All predators	Primary predators only	Secondary predators only
Plants	Top-down ∩ Bottom-up	0.0618	0.0606	0.0000
	Top-down ∩ Other variables	0.2567	0.2454	0.0243
	Bottom-up ∩ Other variables	0.0565	0.0565	0.0565
Herbivores	Top-down ∩ Bottom-up	0.0613	0.0611	0.0233
	Top-down ∩ Other variables	0.2605	0.2588	0.0479
	Bottom-up ∩ Other variables	0.1065	0.1065	0.1065
Planktivores	Top-down ∩ Bottom-up	0.0655	0.0700	0.0075
	Top-down ∩ Other variables	0.4439	0.4183	0.0871
	Bottom-up ∩ Other variables	0.1050	0.1050	0.1050
Herbivores and planktivores	Top-down ∩ Bottom-up	0.0741	0.0698	0.0132
	Top-down ∩ Other variables	0.4064	0.3721	0.0723
	Bottom-up ∩ Other variables	0.1056	0.1056	0.1056

In addition, the results from our cited companion paper (*2*) show that the combination of wave disturbance and El Niño–Southern Oscillation (ENSO) explains only 6% of the variance, and in situ temperature explains less than 1% of the variance in kelp forest community structure based on 18 years of data spanning several strong and weak ENSO events. As we noted (see SOM), the use of satellite-derived productivity data in coastal waters has been extensively validated in our study region [(see also (*3*)]. Importantly, the variation in primary production (*4*) is sufficient to detect potential bottom-up effects, despite missing the extreme nutrient limitation encountered at the southern limit of *M. pyrifera*.

Other variables such as geographic location are, indeed, at least as important as the top-down variables we identified (*2*). However, explaining 20% of variation in community structure is a notable result (*5*), and these "other" variables are largely outside the human influence and so less useful for management purposes.

The claim that our results run counter to the literature on kelp forest ecology is untrue [see, for example, (*6*–*9*)], and we disagree with the suggestion that bottom-up effects offer a more parsimonious explanation of our results. Also, the referenced bottom-up associations are not tests of nutrient versus predator effects on entire kelp forest communities and counter examples exist, as with the monitoring of extreme eutrophication of kelp forests off San Diego that found no effect on kelp forest communities (*10*). Consequently, compensatory mechanisms among species are likely more important than a simple trophic cascade framework would suggest, with these effects driven by top-down forces. Our novel approach allowed us to uncover these results and to open up the quest for the mechanisms driving them.

BENJAMIN S. HALPERN,[1] KARL COTTENIE,[2] BERNARDO R. BROITMAN[1]

[1]National Center for Ecological Analysis and Synthesis, University of California, Santa Barbara, CA 93101, USA. [2]Department of Integrative Biology, University of Guelph, Guelph, ON N1G 2W1, Canada.

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CORRECTIONS AND CLARIFICATIONS

Science Careers: "Young scientists need firm plan to make up for a late start" by K. Robinson (8 Sept., p. 1454). Several lines near the end of the article on page 1457 were dropped during production. The correct passage should read:

"Don't assume that your 403(b) plan representative can help you. They may be trained in sales, not financial planning, and may not know, for instance, whether you have enough emergency cash set aside. You will be served best by an adviser who will consider not just investments but all aspects of your financial life.

Consumer advocates—including Consumers Union, the nonprofit publisher of Consumer Reports—recommend a 'fee-only' financial adviser: one who takes no commissions and is paid directly by the client."

News of the Week: "Genomes highlight plant pathogens' powerful arsenal" by E. Stokstad (1 Sept., p. 1217). The photo credit should be "D. Schmidt, Garbelotto Laboratory, UC Berkeley."

News Focus: "One year after, New Orleans researchers struggle to rebuild" by J. Kaiser (25 Aug., p. 1038). The statement "New enrollment at Tulane's medical school is down by about one-third" refers specifically to graduate students. First-year medical student enrollment is 165 this year, 10 more than in previous years.

Policy Forum: "Public acceptance of evolution" by J. D. Miller *et al.* (11 Aug., p. 765). The URL for the Supporting Online Material is incorrect. It should be www.sciencemag.org/cgi/content/full/313/5788/765/DC1. The link has been corrected in the online version.

Reports: "Permanent El Niño–like conditions during the Pliocene warm period" by M. W. Wara *et al.* (29 July 2005, p. 758). In references 7, 9, and 10, the first author should be D.-Z. Sun, not D.-E. Sun.

TECHNICAL COMMENT ABSTRACTS

Comment on "A Keystone Mutualism Drives Pattern in a Power Function"

David Alonso and Mercedes Pascual

Vandermeer and Perfecto (Reports, 17 February 2006, p. 1000) reported a general power law pattern in the distribution of a common agricultural pest. However, there is an exact analytical solution for the expected cluster distribution under the proposed null model of density-independent growth in a patchy landscape. Reanalysis of the data shows that the system is not in a critical state but confirms the importance of a mutualism.

Full text at www.sciencemag.org/cgi/content/full/313/5794/1739b

Comment on "A Keystone Mutualism Drives Pattern in a Power Function"

Salvador Pueyo and Roger Jovani

Vandermeer and Perfecto (Reports, 17 February 2006, p. 1000) maintain that a mutualist ant disrupts the power law distribution of scale insect abundances. However, reanalysis of the data reveals that ants cause an increase in the range of the power law and modify its exponent. We present a tentative, but more realistic, model that is suitable for quantitative predictions.

Full text at www.sciencemag.org/cgi/content/full/313/5794/1739c

Response to Comments on "A Keystone Mutualism Drives Pattern in a Power Function"

John Vandermeer and Ivette Perfecto

The comments by Alonso and Pascual and by Pueyo and Jovani clarify the power law distribution of subpopulations of the scale insect *Coccus viridis*. The low density deviations are now seen as part of a negative binomial distribution and the high density deviations as resulting from a change in the parameters of the power law. Our biological conclusion that an ant mutualism modifies the form of the power law is thus strengthened.

Full text at www.sciencemag.org/cgi/content/full/313/5794/1739d

Letters to the Editor

Letters (~300 words) discuss material published in *Science* in the previous 6 months or issues of general interest. They can be submitted through the Web (www.submit2science.org) or by regular mail (1200 New York Ave., NW, Washington, DC 20005, USA). Letters are not acknowledged upon receipt, nor are authors generally consulted before publication. Whether published in full or in part, letters are subject to editing for clarity and space.

PUBLIC HEALTH

From Miasmas to Microbes

Hugh Pennington

Cleansing rituals after defecation are universal among humankind. They differ in detail in different cultures, but in essence are the same, because their driving force everywhere is disgust. Our personal reactions to the vomit, pus, slime, and sputum of others are no different. They extend to the products of putrefaction as well. So deeply embedded are these feelings of revulsion and so widespread are they that it is difficult to disagree with those who propose a Darwinian origin for them—that they are the product of an evolutionary process driven by the selective force of infections transmitted by foulness (*1*).

The Great Stink of Paris and the Nineteenth-Century Struggle Against Filth and Germs

by David S. Barnes

Johns Hopkins University Press, Baltimore, MD, 2006. 328 pp. $35, £23.50. ISBN 0-8018-8349-0.

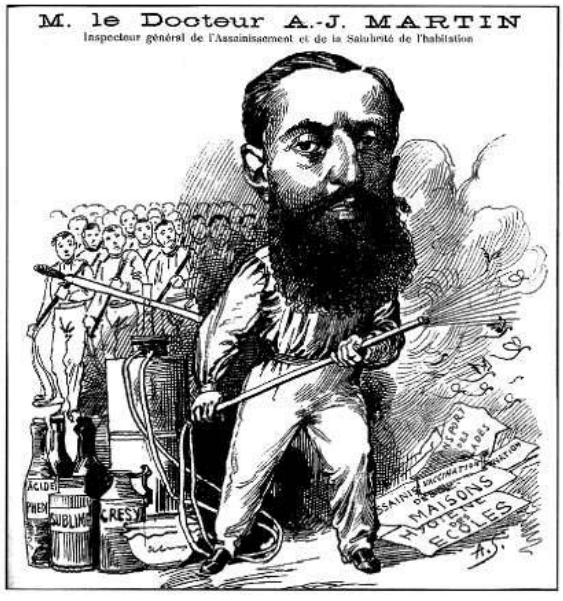
For most of recorded history, the popular view that such things are dangerous to health has had professional medical support. "Airs, Waters, Places," written by Hippocrates 2500 years ago, is the foundation text. A particularly vigorous and influential miasmatist in the 19th century was Florence Nightingale. She said that "the very first canon of nursing … the first essential to the patient, without which all the rest you can do for him is as nothing … is this: TO KEEP THE AIR HE BREATHES AS PURE AS THE EXTERNAL AIR." Foul air was the most important cause of infection: "of the fatal effects of the effluvia from excreta it would seem unnecessary to speak were they not so constantly neglected" (*2*).

However, even as Nightingale's work in the Crimean War of 1854–56 was making her a public figure, the case was being built that other transmission routes for infection were the really important ones. For his conclusions on the spread of cholera, John Snow is seen today as a mid-19th-century hero. The removal, at his instigation, of the handle of the Broad Street pump in Soho in London on 8 September 1854 has caused him to be canonized in recent times as the original shoe-leather epidemiologist, because it is said that his action brought the epidemic to an end. Saints have to be above suspicion. Snow's life was exemplary. But mythologists have been at work as well (*3*). The epidemic had run out of steam well before the handle was removed. And many years went past before his hypothesis that cholera was water-borne received universal consensus. There is a general principal at work here. It is that in public health matters, nothing is pure, neither is it simple. That is a theme that runs through David Barnes's *The Great Stink of Paris and the Nineteenth-Century Struggle Against Filth and Germs.*

Barnes (a historian of science at the University of Pennsylvania) starts his account with "*des emanations odorants de Paris*" of the summer of 1880 and finishes with another bad smell episode in the city 15 years later. Because of the outrage and fear of plagues that the first caused, it led to the establishment of a national commission. In 1895, there was no such response; the bacteriological revolution, the clear association of specific diseases with specific cultivatable microbes, was now well under way. But Barnes's thesis is that the clash of the old-fashioned miasmatic theories and the developing new science did not lead to the defeat of the former and victory for the latter. Rather, as he says, "filth and germs came to be conjoined." Thus his account is not about a hardy band of inspired researchers vanquishing the resistance of hide-bound traditionalists and bringing the light of science at long last to the study of disease. Rather, it discusses the development of what Barnes calls the "sanitary-bacteriological synthesis," which "brought the commonsense cultural appeal and broad applicability of the old knowledge (for example, that foul-smelling substances are bad for one's health) into harmony with the specificity and scientific mastery inherent in the new knowledge of microbes."

Miasmas were replaced by microbes, but feces were still in the frame. Barnes's detailed and scholarly account is persuasive. Many will be surprised to learn about the fondness of Parisian householders for their cesspits, emptied into carts by *vidangeurs* (and sometimes, nocturnally into gutters in the street)—a fondness matched by their resistance to main drainage and "*tout-à-l'égout*" ("everything into the sewers"), first proposed in the mid-1870s. Only in 1903 did houses in Paris with direct sewer connections begin to outnumber those with cesspits; there were still more than 25,000 of the latter on the eve of the First World War. Rather than the 630 kilometers of Baron Haussmann's new Second Empire sewers being a solution, many Parisians saw them as the source of the smell.

But this is France, and its exceptionalism and that of Paris is beyond dispute. Those familiar with French history will have memories jogged appropriately by the author's references to the Franco-Prussian War, the siege of Paris, and the Commune. Nevertheless, the references are too brief. More detail would not only have lightened what is for the general reader a rather serious account, but would have put the 1880 Stink into the necessary context of the national humiliation of defeat by the Germans in 1870–1871 (as the French Corps Commander General Ducrot said at Sedan in August 1870, "*Nous sommes dans un pot de chambre et nous y serons emmerdes!*"), the concentrated and anachronistic nature of Paris, and the shortage of plumbers and other artisans that persisted for years after the mass executions of Communards (at least 20,000 were summarily shot) in the early summer of 1871. These events were still fresh in the memory in 1880, when an amnesty came in for surviving Communards. And Louis Pasteur, a member of the stink commission, refused to use the term "bacteriology," because he considered it to be a constricting "Teutonic" label. In



Clearing the air. Dr. André-Justin Martin led the municipal disinfection service in 1890s Paris.

CREDIT: FROM THE NEWSPAPER *LA VILLE*, 18 NOVEMBER 1893

The reviewer is at the University of Aberdeen Medical School, Polwarth Building, Aberdeen, AB25 2ZD, UK. E-mail: mmb036@abdn.ac.uk

1871, he vowed to inscribe all his publications with the words "Hatred toward Prussia. Revenge! Revenge!" and he returned the honorary MD degree that he had received from the University of Bonn in 1868 (*4*).

But these omissions do not diminish the significance of Barnes's analysis. His sanitary-bacteriological synthesis—"the lasting legacy of the sanitarians of the early nineteenth century, the bacteriologists of the late nineteenth century, and the many preachers of the gospel of germs who have done missionary work at home and abroad ever since"—is very much alive and well in our own time. Cleanliness has never been as close to godliness (and good health) as it is today, thanks as much to our Darwinian disgust of dirt as to science.

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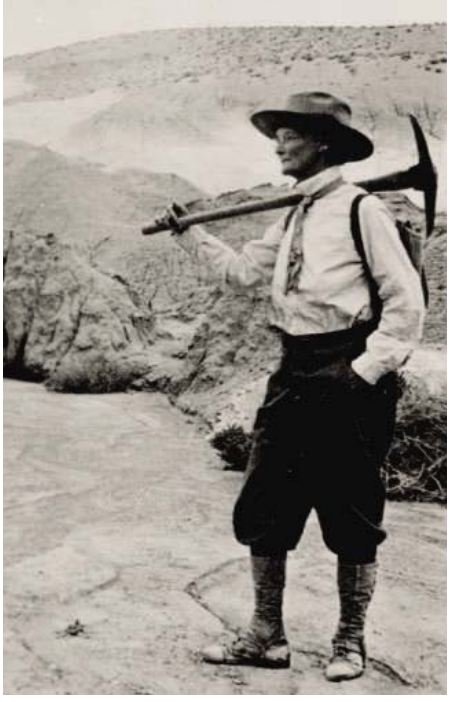

Collectors' benefactress. After founding both Berkeley's Museum of Vertebrate Zoology and the University of California Museum of Paleontology, naturalist Annie Montague Alexander supported them for more than 40 years.

HISTORY OF SCIENCE

Saving American Naturalists from Oblivion

Peder Anker

There are few naturalists and collectors in the pantheon of science, at least if one is to judge by brand-name recognition. Yet the environmental debates of today would have been impossible without their meticulous work. In *All Creatures: Naturalists, Collectors, and Biodiversity, 1850–1950*, Robert E. Kohler seeks to save some of them from oblivion.

All Creatures
Naturalists, Collectors, and Biodiversity, 1850–1950

by Robert E. Kohler

Princeton University Press, Princeton, NJ, 2006. 379 pp. $35, £22.95. ISBN 0-691-12539-2.

Kohler, a prize-winning historian of science at the University of Pennsylvania, may be familiar to readers of *Science*. His earlier *Lords of the Fly* (*1*) brought laboratory history to a new level, and the more recent *Landscapes and Labscapes* (*2*) shows how laboratory ideals came to shape the science of ecology. In this new, well-argued book, Kohler plays down the importance of laboratory life to naturalists. Instead he puts their scientific achievements into the contexts of the environment they worked in, the social culture of nature-going they often came from, and, lastly, the science of classification in the tradition of the Swedish naturalist Carl von Linné.

The book's title should not be taken too literally, as Kohler only tells the story of collecting, describing, and cataloging "all creatures" in the U.S. environment. The word "biodiversity" is also slightly misplaced, as the term was not in use until the late 1980s. Kohler's focus is on the life and work of naturalists of the late 19th century. Instead of telling their stories from a beginning to an end, as historians usually do, Kohler has organized the book according to a set of conditions that came to shape their natural surveys. In doing so, he avoids lines of arguments that could inflame the now-expiring science war of the late 1990s. Kohler argues that both external and internal factors explain scientific discoveries and developments: "Environment, culture, and science in concert" produced the age of natural surveys.

Kohler offers an innovative reading of turn-of-the-century American landscapes as "inner frontiers." At this time, he argues, U.S. pioneers could no longer expect to find a linear border of unsettled land in the West; instead they encountered a mosaic of settled and uninhabited areas. This landscape of inner frontiers provided naturalists with a unique combination of wildness and accessibility. "The ecologist Arthur Vestal was amazed to find a patch of original California prairie in the vacant lot just a few doors from his home in Stanford," for example. New roads and rail tracks also made previously hard-to-get-to places within easy reach. It was this unique historical situation, Kohler contends, that made possible large-scale scientific surveys of U.S. species.

Equally important was the culture of outdoor recreation, camping, and back-to-nature lifestyles. Many naturalists found their own passion for nature in this milieu, and it was also among nature-goers that they found patronage. The audience of large natural history museums wanted to see wild creatures on display, and the naturalists were sent out to collect them. Kohler provides a rich account of these and similar social circumstances that made grand natural surveys possible.

Out in the field the collectors had to proceed according to certain methodological procedures that were anything but easy. They also faced social obstacles working in environments that were used by varied sorts of people. Being recognized as field scientists was not a matter of course, as naturalists often were perceived as odd strangers by outsiders: e.g., "C. Hart Merriam [the founder of the U.S. Biological Survey] was mistaken for a bank robber hiding out," and ecologist "Charles C. Adams was taken for a detective, a deadbeat, and a crazy man" when collecting snails in Tennessee. The inclusion of such details makes the book an attractive account of the daily life of naturalists.

All Creatures is intended for "curious general readers" and is written in an engaging language free of technical jargon. Nonetheless, professional historians of various scientific disciplines will also find many topics of interest: Kohler asserts, for example, that scientific developments can be "cyclic or self-limiting," and he thus challenges linear historiography. The book will therefore reward the target audience and specialists alike. An important contribution to the history of naturalists in the United States, it is well worth the read.

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The reviewer is at the Forum for University History, University of Oslo, Post Office Box 1008 Blindern, 0315 Oslo, Norway. E-mail: anker@ffu.uio.no

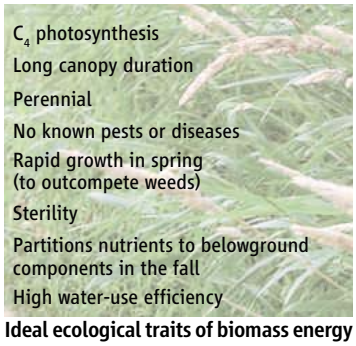
ECOLOGY

Adding Biofuels to the Invasive Species Fire?

Biofuel crops, particularly using non-native species, must be introduced with an understanding of possible risks to the environment.

S. Raghu,[1]* R. C. Anderson,[2] C. C. Daehler,[3] A. S. Davis,[4] R. N. Wiedenmann,[5] D. Simberloff,[6] R. N. Mack[7]

The U.S. renewable energy initiative (*1*) announced in the 2006 presidential State of the Union address (*2*) has given new impetus to the identification of biofuel crops as sources of energy. However, an earlier presidential directive, Executive Order 13112 (*3*), attempts to protect the United States from invasive species, unless benefits clearly outweigh potential harms. The policies may conflict because traits deemed ideal in a bioenergy crop are also commonly found among invasive species (see figure).



Ideal ecological traits of biomass energy crops (*4*). All traits shown other than perennial growth and sterile seeds are known to contribute to invasiveness. See (*25*).

Biofuel crops may have economic benefits, but studies of concomitant environmental risks of movement into novel habitats are seldom conducted. Although anecdotal claims of "low risk" for some species (*4*) may be valid, many purportedly beneficial introduced species have had long-term economic and environmental costs owing to their invasiveness (*5*, *6*). For example, *Sorghum halepense* is an introduced forage grass that became an invasive weed in 16 of the 48 U.S. states in which it occurs. Even the most conservative estimate of competitive losses for cotton and soybean crops in three states is in excess of $30 million annually (*7*).

Several grasses and woody species have been evaluated for biofuel production, with perennial rhizomatous grasses showing the most economic promise (*4*, *8*). *Arundo donax* (giant reed; native to Asia) and *Phalaris arundinacea* (reed canary grass; native to temperate Europe, Asia, and North America) are two C_3 grasses being considered as biofuel species (*8*) that are invasive in some U.S. ecosystems. The former threatens riparian areas and alters fire cycles (*9*); the latter invade wetlands (*10*) and affect wildlife habitat.

The hybrid grass *Miscanthus* × *giganteus* (native to Asia) and *Panicum virgatum* (switchgrass; native to central and eastern United States) are C_4 grasses being considered in Europe and the United States (*4*, *11*). Several *Miscanthus* species are invasive or have invasive potential (*12*); in particular, the parent species of *M.* × *giganteus* (*13*, *14*). *Miscanthus* × *giganteus* is an allopolyploid that does not produce viable seed and reproduces vegetatively. However, allopolyploidy does not guarantee continued sterility (*15*) and vegetative propagation is often associated with invasiveness (*16*, *17*) or directly contributes to it (*18*). Several other traits that make *Miscanthus* potentially valuable as a crop could enhance invasiveness (ability to resprout from rhizomes, efficient photosynthetic mechanisms, and rapid growth rates) (*16*, *19*).

The U.S. native, *P. virgatum*, shares many traits with *Miscanthus* and can also produce seeds, which may give *P. virgatum* even greater invasive potential. Furthermore, plants native in one region can become invasive when established elsewhere (*20*). Escape from competitors and natural enemies may help explain the weedy nature of *P. virgatum* outside its endemic range (*21*).

Internationally, there has been little success in eradicating or even controlling an invading grass. Herbicides are used to control invasive grasses on croplands, but they are too expensive to use on rangelands, national parks, and reserves. Development of the most economical tool, biological control with a specific natural enemy, has been avoided because of the perceived risk of its expanding its host range to include commercial grasses, such as wheat, corn, barley, or rice (*22*).

Balancing costs and benefits of species introductions is a key contemporary challenge. Introducing some plant species as biofuel sources may be safe, but safety must be established by agronomic and ecological analyses. Such analyses are already mandatory for biological control agents (*23*) and transgenic plants (*24*). Experts must assess ecological risks before introducing biofuel crops, to ensure that we do not add biofuels to the already raging invasive species fire.

[1]Illinois Natural History Survey, Champaign, IL 61820; [2]Illinois State University, Normal, IL 61709; [3]University of Hawaii at Manoa, Honolulu, HI 96822; [4]USDA-ARS Invasive Weed Management Unit, Urbana, IL 61801; [5]University of Arkansas, Fayetteville, AR 72701; [6]University of Tennessee, Knoxville, TN 37996; [7]Washington State University, Pullman, WA 99164, USA.

*Author for correspondence. E-mail: raghu@uiuc.edu

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Supporting Online Material

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PLANETARY SCIENCE

Meteorites and their Parent Asteroids

Bodies in our solar system have different oxygen isotope signatures, which have now been used to clarify that a collision involving the asteroid Vesta led to two major groups of meteorites.

Robert N. Clayton

Meteorites provide the best available record of the chemical and physical processes that occurred during the first few million years of our solar system's history. However, they are a very imperfect sample in that each meteorite is a small sample of a much larger parent body, probably an unidentified asteroid. It is, therefore, a major challenge to read the record in any individual meteorite or class of meteorites. Fortunately, there are a few meteorite parent bodies that are known and can be studied directly, thus providing a link between observations on vastly different scales. For example, the Moon has been studied extensively by spacecraft and has been sampled directly at a few locations. These results provide a framework for interpretation of lunar meteorites, which originate from a much greater fraction of the lunar surface than has been actually sampled. Similarly, the SNC meteorites (consisting of shergottites, nakhlites, and chassignites, designations that reflect different proportions of the major silicate minerals: feldspar, pyroxene, and olivine), probably derived from Mars, provide information that is complementary to that obtained by surface analyses and remote sensing.

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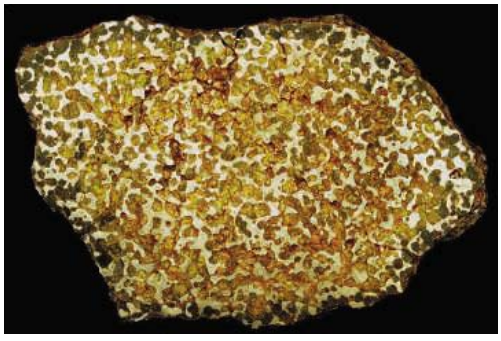
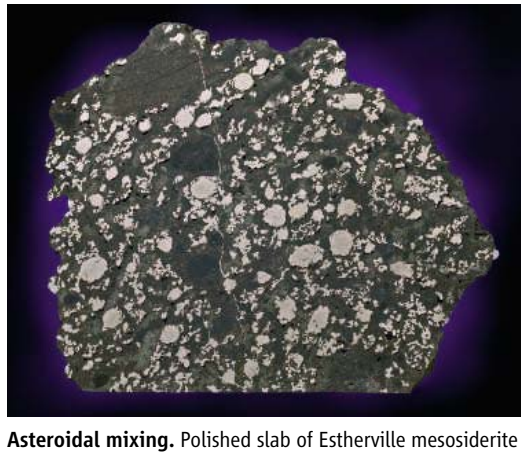
The only other example of a well-established genetic association between a parent body and a meteorite class is that between the asteroid 4 Vesta and the HED meteorites (howardites, eucrites, and diogenites). Vesta is a differentiated body with basaltic rocks at the surface and, presumably, olivine-rich rocks in the mantle and metal-rich rocks in the core, as is found on Earth. On page 1763, Greenwood *et al.* (*1*) seek to identify candidates for metal-rich interior samples of Vesta among the various stony-iron meteorite groups and thus better sort out the parent-child relations among these complex planetary objects. Identification of particular meteorite types with a specific asteroidal source is almost the equivalent of a sample-return space mission, in that it allows precise earth-based laboratory studies of extraterrestrial materials from a known part of the solar system.



Inside an asteroid. Polished slab of Brenham pallasite that was found in 1882 in Kiowa County, Kansas. The specimen (30 cm across) consists of yellowish olivine crystals embedded in iron-nickel metal. Pallasites may represent samples of the core-mantle boundary of an asteroid. Main-group pallasites, such as Brenham, may be from the same body as the Group IIIAB iron meteorites.

The various planetary and asteroidal bodies in the solar system contain a specific isotopic label, implanted at birth and not subsequently modified by interior processes, such as melting, metamorphism, or aqueous alteration. This label is in the form of characteristic abundances of the three stable isotopes of oxygen, the most abundant element in rocky bodies. The oxygen isotopic abundances are useful in assigning meteorites to classes representing common source materials (*2*). The oxygen isotope "fingerprint" has been used to support the assignment of the HED meteorites to a single parent body (*3*). The isotopic label also allows distinction among similar lithologies from different parent bodies [e.g. the meteorite Ibitira, long thought to be a member of the eucrite group, is now recognized, on the basis of oxygen isotopes (*4*), to be from a separate parent asteroid]. A previous oxygen isotope study of achondritic meteorites (rocks that were once molten) (*3*) led to the suggestion that the mesosiderites and pallasites may have come from the HED parent body. Pallasites are commonly displayed in museums because of their spectacular appearance—coarse olivine crystals "floating" in a sea of metal—and have often been described as a transition layer between a metallic core and a rocky mantle (see the first figure). Mesosiderites, also mixtures of roughly equal amounts of silicate and metal, generally have a fine-grained fragmental texture, suggestive of an origin by violent mixing in an asteroidal collision (see the second figure).

The new data of Greenwood *et al.* (*1*), with improved analytical precision, are still consistent with a Vesta origin for the mesosiderites but clearly require a different parent body for the main-group pallasites. Both chemical and isotopic evidence implies a genetic association between these pallasites and the iron meteorites of group IIIAB, a presumed planetesimal core, but this isotopic signature is not known in any stony meteorites that might represent the mantle of the planetesimal. Their study goes beyond identifying a source for the mesosiderites: These meteorites are complex, fine-grained rocks with roughly equal amounts of metal and silicates, so that their structure has major implications for the collisional history of planetary bodies.



Asteroidal mixing. Polished slab of Estherville mesosiderite that fell in 1879 in Emmet County, Iowa. The specimen (24 cm across) consists of numerous rounded metal nodules (several centimeters in diameter) and large, angular silicate clasts (up to 8 cm in diameter) enclosed in a finer grained matrix of silicate-rich material and irregularly shaped metal-rich patches. A number of the silicate clasts are cross-cut by thin, late-stage metal veinlets. Estherville, like the other mesosiderites, is believed to have formed when the molten metal-rich core of an asteroid, from which the outer crust and mantle had been removed, impacted and mixed with the relatively cold silicate-rich, surface layers of a second differentiated asteroid.

The author is at the Enrico Fermi Institute, University of Chicago, Chicago, IL 60637, USA. E-mail: r-clayton@uchicago.edu

Greenwood *et al*. (*1*) demonstrate the utility of the oxygen isotope signature in matching up meteorites to parent bodies but do not specifically address the unsolved question of the origin of these signatures. In simplest terms, the isotopic signature can be expressed by the quantity $\Delta^{17}O$, defined as $\Delta^{17}O = \delta^{17}O - 0.52\delta^{18}O$, where $\delta^{x}O = 1000\{[(^{x}O/^{16}O)_{sample}/(^{x}O/^{16}O)_{standard}] - 1\}$. $\Delta^{17}O$ is the vertical displacement of a data point on a graph of $\delta^{17}O$ versus $\delta^{18}O$, relative to the terrestrial mass–dependent fractionation line, for which $\Delta^{17}O \equiv 0$. This quantity is the ordinate in figure 1 of Greenwood *et al.* (*1*), which shows three horizontal lines corresponding to (i) $\Delta^{17}O = 0$ (the terrestrial fractionation line, TFL), (ii) $\Delta^{17}O = -0.183$ (pallasites), and (iii) $\Delta^{17}O = -0.245 \pm 0.20$ (mesosiderites). A previous study (*5*) found $\Delta^{17}O = -0.239 \pm 0.007$ for the HED group. For the SNC meteorites (from Mars), $\Delta^{17}O = 0.321 \pm 0.013$ (*6*). Thus, $\Delta^{17}O$, defined as zero for Earth, has a negative value for the asteroid Vesta and a positive value for the planet Mars, which clearly is at odds with a commonly made assumption of a monotonic variation of $\Delta^{17}O$ with radial distance from the Sun.

Another outstanding problem in the use of oxygen isotopes to understand planetary accretion is the exact coincidence in isotopic composition between Earth and the Moon (*7*). In light of the conclusions in the previous paragraph, there is no justification for the statement that the proto-Earth and the Moon-forming impacts were formed "at about the same heliocentric distance" (*7*). A more fruitful approach may be one that involves extensive material exchange between target and impactor (*8*), so that previous isotopic differences were erased.

An additional unsolved problem in planet formation is the possibility of large oxygen isotope differences between the Sun and the inner planets. In contrast to the very small isotopic differences among the large, differentiated bodies, the differences in isotope ratios among small, primitive meteoritic objects (such as chondrules and refractory inclusions) are several percent, rather than <0.1% (*2*). The origin of these variations is thought to be an isotope-selective photodissociation of carbon monoxide in the solar nebula (*9*–*11*). These models predict that the isotopic ratios, $^{17}O/^{16}O$ and $^{18}O/^{16}O$, in the nebular gas and in the present-day Sun are about 5% lower than those ratios in oxygen of the solid Earth and other inner planets. It remains unclear how the planets achieved such an unusual isotopic composition and yet are very similar to one another. Measurement of the oxygen isotopic composition of the Sun is the highest priority of NASA's Genesis mission (*12*). Another planned NASA mission (Dawn) will orbit the large asteroids, Vesta and Ceres, and will provide observational tests of the proposals of Greenwood *et al*. (*1*) concerning rock types on Vesta.

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NEUROSCIENCE

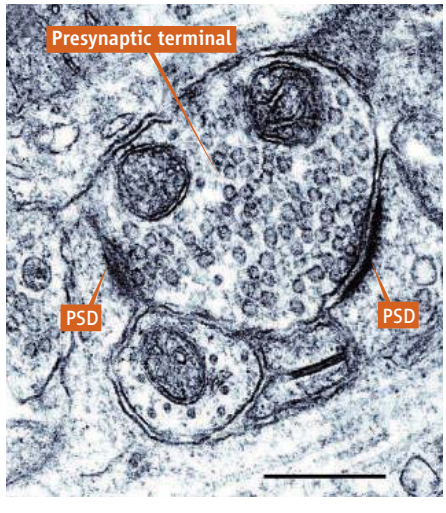
Adam Finds an Exciting Mate

Solomon H. Snyder

Although a principal focus of epilepsy research has been on ion channels, a ligand-receptor interaction in neurons may also be important in the disease.

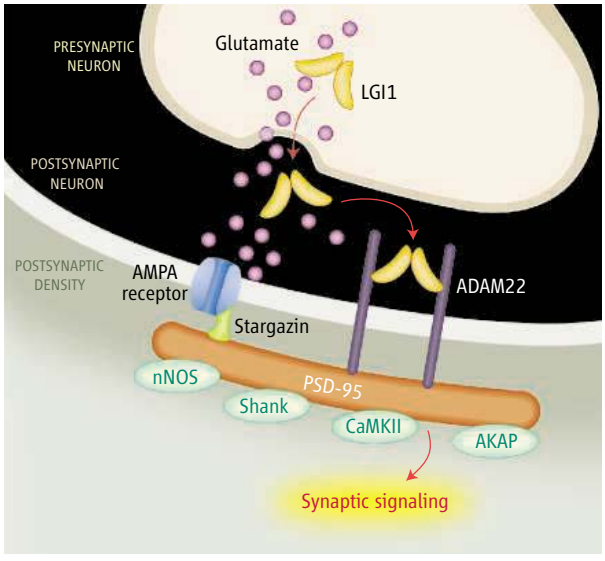
Epilepsy, one of the most common neurological disorders, is caused by abnormal electrical activity in the brain. The symptoms range in severity from mild sensory disruption to recurring seizures and unconsciousness. Most forms of epilepsy have been assumed to stem from brain tissue "scars" acquired through trauma, so that molecular approaches to understanding and treating the disease would be fruitless. In recent years, mutations in various ion channels have been linked to rare forms of epilepsy. Now, on page 1792 in this issue, Fukata *et al*. (*1*) identify a pairing of proteins in neurons that may be relevant to the pathogenesis of human epilepsy. The agonist-receptor pair regulates the activity of the glutamate-AMPA (α-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid) receptor, the subtype that mediates the most prominent form of excitatory neurotransmission in the brain.

At the cleft, or synapse, between two communicating neurons lies the postsynaptic density, the region in the postsynaptic neuron where adhesion and signaling molecules are organized to facilitate rapid response to neurotransmitter molecules released by the presynaptic neuron (see the first figure). Key among these constituent molecules is postsynaptic density-95 (PSD-95), a major scaffolding protein localized to the postsynaptic density of brain synapses. Fukata *et al*. sought binding partners for PSD-95 by monitoring proteins that associate with it in rat brain extracts. The authors identified a protein complex comprising stargazin, LGI1, and ADAM22 (see the second figure). Interestingly, all three proteins have previously been implicated in epilepsy. LGI1 is a known secreted neuronal protein, which is mutated in patients with a rare genetically determined form of epilepsy but whose physiological role has been obscure (*2*). ADAM22 is a neuronal membrane protein. Its mutation in mice leads to death from seizures, but its normal function has also not been definitively established (*3*). Stargazin,



Where information is processed. Electron micrograph shows postsynaptic densities (PSD) in the postsynaptic membranes of dendritic spines where a presynaptic nerve terminal forms synapses. Scale bar, 400 nm (*6*).

The author is in the Department of Neuroscience, Johns Hopkins University School of Medicine, 725 North Wolfe Street, Baltimore, MD 21205, USA. E-mail: ssnyder@jhmi.edu



Synaptic transmission and epilepsy. LGI1, a secreted protein presumably released by the presynaptic neuron, binds ADAM22 on the surface of the postsynaptic neuron, increasing synaptic signal transmission. This interaction may be linked to some forms of epilepsy. LGI1 is an oligomer that binds to two molecules of ADAM22. The intracellular portion of ADAM22 binds PSD-95, a scaffolding protein that is associated with AMPA receptors and multiple signaling proteins. Some of these proteins could modulate LGI1-ADAM22 interaction. nNOS, neuronal nitric oxide synthase; CaMKII, calcium-calmodulin kinase II; AKAP, A-kinase anchoring protein.

discovered as the protein that is mutated in Stargazer mice that suffer from epilepsy, is a subunit of the AMPA receptor and regulates AMPA receptor trafficking to the postsynaptic density (*4*). Fukata *et al*. establish that LGI1 binds selectively to ADAM22, specifically at synaptic sites on the surface of rat brain neurons. Treatment of rat brain slices with LGI1 augmented AMPA receptor–mediated neurotransmission, reflected by the increased amplitude and frequency of postsynaptic AMPA receptor–meditated electric currents. This action was prevented by first treating the brain slices with a soluble form of ADAM22, indicating that synaptic activation by LGI1 is dependent on its binding to ADAM22. The increased neurotransmission appears to reflect recruitment of new AMPA receptors to the postsynaptic density, because LGI1 expression in cultured hippocampal neurons increases surface expression of AMPA receptors.

How do these findings influence our understanding of excitatory neurotransmission in the brain? AMPA receptor–mediated neurotransmission is dynamically regulated by trafficking of the receptor. A variety of proteins bind directly or indirectly to the receptor, down-regulating transmission by causing internalization of the receptors or augmenting transmission by increasing surface expression. Such proteins include shank, NSF, GRIP, and PICK-1, all of which are localized in the postsynaptic neuron (*5*). The LGI1-ADAM22 connection appears to be the first example of a receptor-regulating protein that is released by neurons, likely from their synaptic terminals, and binds to a partner, ADAM22, on the external surface of the postsynaptic membrane. Although neurotransmitters are generally small molecules, relatively large-sized LGI1 (about 60 kD) appears to serve a neuromodulatory function as well. It is not likely that LGI1 would be regarded as a putative neurotransmitter. However, its presumed release from nerve terminals and actions at postsynaptic membrane receptors raise questions about the definition of "neuromodulation" and "neurotransmission." Proteins as large as insulin have been proposed as neurotransmitters.

Thus far, little is known about LGI1 biosynthesis, degradation, and release. If these processes are dynamically regulated by depolarization of the presynaptic neuron, LGI1 may well emerge as a major determinant of brain excitation. Similarly, the function of ADAM22 has hitherto been obscure, but it also appears to physiologically influence AMPA receptor-mediated neurotransmission. How ADAM22 interacts with the other PSD-95–binding proteins, such as nitric oxide synthase, stargazin, shank, calcium-calmodulin kinase II, and A-kinase anchoring protein, remains to be determined. Perhaps these other proteins mediate the effects of ADAM22 on AMPA receptor expression and function.

The generation of epileptic seizures has usually been ascribed to changes in neuronal ion channels. The finding that mutation of various proteins associated with AMPA receptors leads to epilepsy suggests that glutamate neurotransmission plays a more prominent pathogenic role than previously appreciated. The postsynaptic density, a structure comprising several scaffolding proteins linked to AMPA receptors, might be disrupted by the "scarring" that underlies many forms of epilepsy. Conceivably, hitherto undetected mutations in the proteins of postsynaptic densities may be responsible for multiple forms of seizure disorders.

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MOLECULAR BIOLOGY

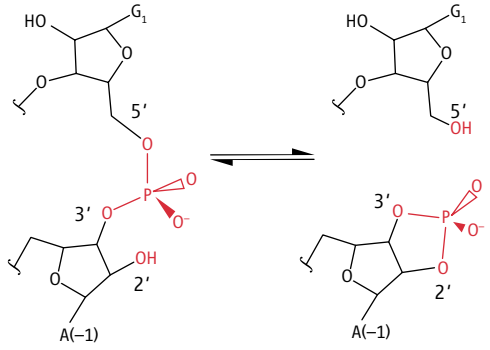
Versatility of Self-Cleaving Ribozymes

Michael D. Been

Two studies expand our knowledge of cellular RNA sequences that catalyze their own cleavage. One sheds light on their evolution, the other on the cleavage mechanism.

Short RNA sequences that catalyze their own cleavage (self-cleaving ribozymes) are sometimes viewed as descendants of an early, even pre-protein, era of life, because they are made of genetic material but possess enzyme-like properties. Most self-cleaving ribozymes are associated with exotic small replicating RNAs called replicons, many of which do not code for proteins (*1*). Recently, however, sequences for self-cleaving ribozymes have been found associated with protein-coding cellular genes. These ribozymes are thought to regulate or modify gene expression. The new discoveries raise questions about how these potentially more "modern" ribozymes may differ from those that replicate RNA. Moreover, perhaps our timetable is wrong; self-cleaving ribozymes associated with gene regulation may have predated and have been the ancestors of the self-cleaving sequences in some small virus-like replicons.

The author is in the Department of Biochemistry, Duke University Medical School, Durham, NC 27710, USA. E-mail: mdbeen@duke.edu



Cleaving the RNA backbone. The reactions catalyzed by self-cleaving ribozymes generate a 2′,3′ cyclic phosphate, which cleaves the 3′-5′ phosphodiester backbone in the RNA strand. The *glmS* ribozyme also uses this mechanism, but unlike other small ribozymes, a metabolic intermediate required for cell growth triggers the cleavage reaction.

Two reports in this issue refine our picture of self-cleaving ribozymes associated with protein-coding genes. Klein and Ferré-D'Amaré [page 1752, (*2*)] zoom in and provide a sharp image of the active site of a bacterial self-cleaving ribozyme. Salehi-Ashtiani *et al.* [page 1788, (*3*)] pan back and provide a broader view of the possible evolution of self-cleaving ribozymes.

Klein and Ferré-D'Amaré investigate the *glmS* ribozyme, which was discovered by Breaker and co-workers (*4*, *5*) in a computational search for a type of structured RNAs called riboswitches. Most riboswitches regulate gene expression through a structural rearrangement caused by binding to a small metabolite ligand (*6*). In contrast, the glmS riboswitch cleaves itself (see the first figure) in the presence of glucosamine-6-phosphate (GlcN6P), a metabolite required for cell growth (*4*, *5*). The *glmS* ribozyme is the first example of a natural ribozyme that is also a riboswitch.

It has been proposed that the *glmS* ribozyme regulates cellular production of GlcN6P by controlling the amount of the GlcN6P synthase (*4*, *5*). In the *glmS* mRNA transcript, the ribozyme is followed by the RNA coding for the synthase. According to the model, if the amount of GlcN6P exceeds that needed for cell growth, GlcN6P interacts with the ribozyme, the RNA is cleaved, and the segment coding for the synthase is not translated. When the amount of GlcN6P is too low, the ribozyme is inactive, and more synthase is made. The result would be regulation of the rate of synthesis of GlcN6P through its direct interaction with the GlcN6P synthase mRNA.

Biochemical studies have provided insights into how GlcN6P may facilitate RNA cleavage. The ribozyme cleaves fastest in the presence of GlcN6P, but a few small molecules that differ from GlcN6P in shape and size, but have similarly positioned amino and hydroxyl groups, support cleavage at slower rates (*7*). These data, together with evidence that the ribozyme appears not to change its overall shape with addition of GlcN6P (*4*), suggest that the ligand provides a catalytic group for the reaction (*7*).

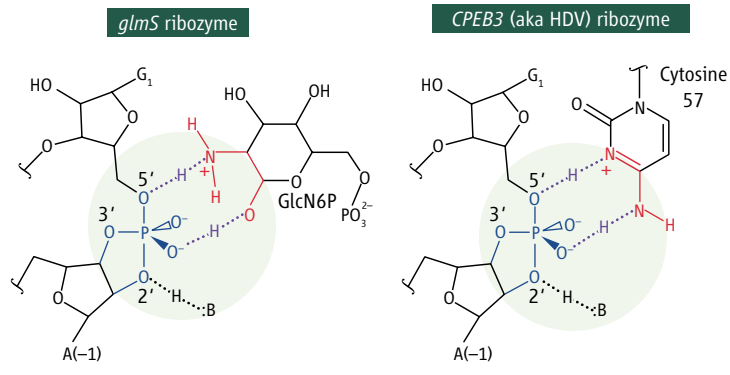
Klein and Ferré-D'Amaré now provide details about how this GlcN6P-dependent ribozyme works. They crystallized the *glmS* ribozyme from *Thermoanaerobacter tengcongensis* in several forms that included a precursor, a precursor bound to a non-activating analog of GlcN6P, and the cleaved product RNA. The ribozyme forms a highly contorted double pseudoknot that positions the cleavage-site phosphate in the active site and generates a solvent-accessible binding pocket for GlcN6P. Potential catalytic groups of a bound ligand are positioned such that they can assist in the reaction (see the second figure, left panel). Other ribozymes may use metal ions as cocatalysts in roles similar to that played by GlcN6P, but the use of a metabolic intermediate appears to be unique to this ribozyme.

Given that the *glmS* ribozyme is present in some pathogenic bacteria (such as *Bacillus anthracis*), could the structures reported by Klein and Ferré-D'Amaré provide the information needed to design a drug targeted to the ribozyme? This challenge could prove to be interesting. Shutting off GlcN6P (and cell-wall) synthesis will require a cocatalyst that binds more tightly than GlcN6P but still catalyzes cleavage to ensure rapid inactivation of the mRNA that codes for the GlcN6P synthase. Even then, the chances of a mutation reducing ribozyme activity and conferring resistance would be high.

Salehi-Ashtiani *et al.* take a different approach to learn more about self-cleaving ribozymes. Their aim was to find self-cleaving ribozyme sequences in the human genome. To identify such sequences, they designed a system that mimicked the replication of small RNA replicons, where newly synthesized RNA spools continuously off a circular template (rolling circle replication) and is cut to the correct size by a self-cleaving ribozyme in the newly made RNA. Unlike the RNA replicons, the templates were small duplex DNA circles that each contained a random fragment (about 150 base pairs) of human DNA and a transcriptional promoter. Rolling circle replication generated continuous head-to-tail repeats of RNA sequences. A tiny fraction of the sequences that cleaved to discrete RNA-repeat sizes were amplified and identified.

The authors found four self-cleaving ribozyme sequences, one of which mapped to the first intron of the cytoplasmic polyadenylation element binding protein 3 (*CPEB3*) gene. (An intron is a DNA sequence that does not code for a protein and is removed before translation of mRNA). An analysis of genomic data revealed that *CPEB3* and the *CPEB3* ribozyme are present in all mammals, but not in other vertebrates. There is evidence that the *CPEB3* ribozyme is active in tissues.

The *CPEB3* ribozyme shows striking resemblance in structure and mechanism to the ribozymes in hepatitis delta virus (HDV) (*8*), a pathogenic virus-like particle found naturally only in humans. The similarity to HDV includes an essential active-site cytosine (C57) (see the second figure, right panel). The *CPEB3* ribozyme cleaves more slowly than the viral ribozyme, but that difference may reflect a requirement for reduced activity to allow gene expres-



Active sites for self-cleaving ribozymes. **(Left)** In the *glmS* ribozyme, the amino group and adjacent hydroxyl group of a bound GlcN6P are within hydrogen-bonding distance to the 5′ bridging oxygen and a nonbridging phosphate oxygen, respectively. This arrangement could increase the rate of cleavage by stabilizing the transition state and, perhaps, catalyzing proton transfer [see figure 6 in (*2*)]. **(Right)** The *CPEB3* ribozyme requires a cytosine at position 57 for cleavage activity [see figure 3 in (*3*)]. It has been proposed (*9*) that in HDV, the equivalent cytosine (*8*) is positioned much like the GlcN6P.

sion. Perhaps harder to explain is why an active self-cleaving ribozyme sequence exists within an intron.

The similarity between the *CPEB3* and HDV ribozymes is unlikely to be coincidental. Could it be that, long after proteins had become the dominant biological catalysts, HDV chose to adopt a ribozyme from the cellular genome?

The two papers will stimulate more studies about the mechanism of self-cleaving ribozyme activity and how they control gene expression. Origin-of-life remnants or not, self-cleaving ribozymes serve diverse and highly specific functions in today's protein-rich cellular world. Given the apparent versatility of self-cleaving ribozymes, future studies are likely to reveal further roles for such ribozymes.

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CHEMISTRY

A Pixellated Window on Chemistry in Solids

Reaction dynamics in solid samples can be studied with a laser beam that has traveled through specially cut windows. Events happening at different times are probed in a single shot.

V. A. Apkarian

Freezing usually arrests chemistry, stopping even the most reactive free radicals from reacting by trapping them in a "cage" in the solid lattice. Yet, some of the most violent reactions occur in solids. An example well known to students is the explosive reaction of nitrogen triiodide (*1*). In a display of showmanship, the teacher rubs the crystalline solid with a feather attached to the end of a long pole to generate a loud bang, caused by the breaking of N–I bonds and the formation of N–N and I–I bonds. This violent solid-state reaction requires the stress applied by the feather to be amplified locally by about 18 orders of magnitude in order to break individual chemical bonds: It is as if a zipper were undone by the action of an earthquake.

The same type of multiscale dynamics is manifest in the sparkles visible when chewing candy that glows in the dark, or in the tiny bursts of light produced when "nipping" sugar into smaller chunks, as first noted by Francis Bacon (*2*). The phenomenon, coined triboluminescence in 1895, is another illustration of the conversion of mechanical friction to electronic excitation—generating electrical discharge and light emission (*3*).

Yet despite the age-old fascination with these phenomena, our understanding of the underlying processes remains far from satisfactory. From the inexorably slow chemistry that accompanies petrification to the violence of shock-sensitive detonation, multiple time and length scales govern solid-state chemistry, posing serious challenges to atomic-scale understanding of these processes. On page 1756 in this issue, Poulin and Nelson (*4*) report a method that overcomes some of these problems by probing molecular dynamics occurring at different times in a single experiment.

The primary experimental difficulty in dissecting solid-state chemistry is the irreversibility of the processes at issue. Consider the liquid-phase counterpart to triboluminescence: the process of sonoluminescence. Despite the relative novelty of this phenomenon, current understanding of sonoluminescence has progressed considerably (*5*). Ultrasonic waves can be converted into electronic energy, and possibly even nuclear fusion (*6*), through cavitation (bubble formation), cavity oscillations, and implosion to generate local temperatures as high as 1 million to 10 million K. Knowledge of the molecular structure and composition of these processes is now sufficiently detailed to find practical application in sonochemistry (*7*).

The rate of elucidation of these related phenomena differs so widely because sonoluminescence measurements can be repeated, whereas triboluminescence experiments are irreversible. A single bubble can repeatedly be driven into sonoluminescence at acoustic rates of 10,000 s^{-1}, resulting in perfectly synchronized picosecond-long flashes from the same sample for millions of cycles. In contrast, little material is left behind when a solid detonates. The measurement cannot be repeated on the same sample.

One way to overcome this difficulty is to carry out measurements on carefully prepared homogeneous plates. To emulate repetition, the plates are translated in a laser beam to expose a fresh spot of nearly identical material for experimentation. This approach has been used successfully to characterize shock-induced chemistry and detonation with nanometer-scale space resolution and picosecond-scale time resolution (*8*). In addition to characterizing temperature and pressure profiles of chemically driven shock fronts that arise during detonation, the chemical transformations that sustain the front can now be studied with nonlinear ultrafast spectroscopies (*9*).

The alternative to investigating a complete process on the multiple time and length scales peculiar to chemistry in solids is the reductionist approach. Under the assumption that a complex process may be reconstructed from its parts, elementary steps are isolated for scrutiny. This approach guides the more extensive body of work in solid-state chemical dynamics. For example, doped rare gas solids have been used to dissect the elementary steps of energy flow within and between molecules, energy deposition in molecular bonds up to breakage, caging, and cage escape and migration of atomic fragments into the lattice (*10*). To allow repetition, these measurements are performed in model systems that are designed to be reversible.

However, understanding the elementary steps in isolation will never be sufficient. It is essential to also carry out measurements of real-world processes, which are invariably irreversible. To this end, techniques that do not require repetition and yield the complete

The author is in the Department of Chemistry, University of California at Irvine, Irvine, CA 92697, USA. E-mail: aapkaria@uci.edu

record of the course of molecular dynamics in a single measurement would be invaluable. In this context, the article by Poulin and Nelson represents a valuable advance.

The authors show how one can obtain—in a single shot—time-sequenced transient absorption spectra of irreversible chemical reactions that are initiated with a laser in a solid sample. To do so they use echelons, which are windows cut in steps to provide stripes of different thickness through which the laser traverses. By crossing two echelons, pixels of square profile are created. Each pixel generates a different time delay, allowing events happening at different times to be probed in a single shot.

Poulin and Nelson use this method to record the caging process of the triiodide ion upon its photodissociation in organic crystals. Caging results in the reformation of the broken bond. The bond reformation proceeds coherently in the tight cages, while there is significant dispersion in the timing of recombination in the looser solids. Although the observed caging process is reversible, discoloration of the irradiated spot indicates that nonreversible structural change also occurs, precluding the repetition of measurements on the same spot. The work also highlights that in condensed-phase systems, linear spectroscopies are not uniquely interpretable: Simulations are essential to connect the one-dimensional spectra to the underlying multidimensional dynamics (*4*).

Poulin and Nelson have successfully put into practice the concept of the pixellated window in time. The use of echelons, however, limits the observation window to the picosecond time range. Clever optics will be required to follow processes on femtosecond to millisecond time scales and thus unravel the multiscale dynamics peculiar to solids; for example, to follow not only the bond breaking, but also the process of permanent discoloration seen in the crystals used in their experiments.

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GEOPHYSICS

Do Earthquakes Rupture Piece by Piece or All Together?

Laboratory measurements are being used to resolve which of two models is better at explaining how Earth's crust ruptures to create earthquakes.

Chris Marone and Eliza Richardson

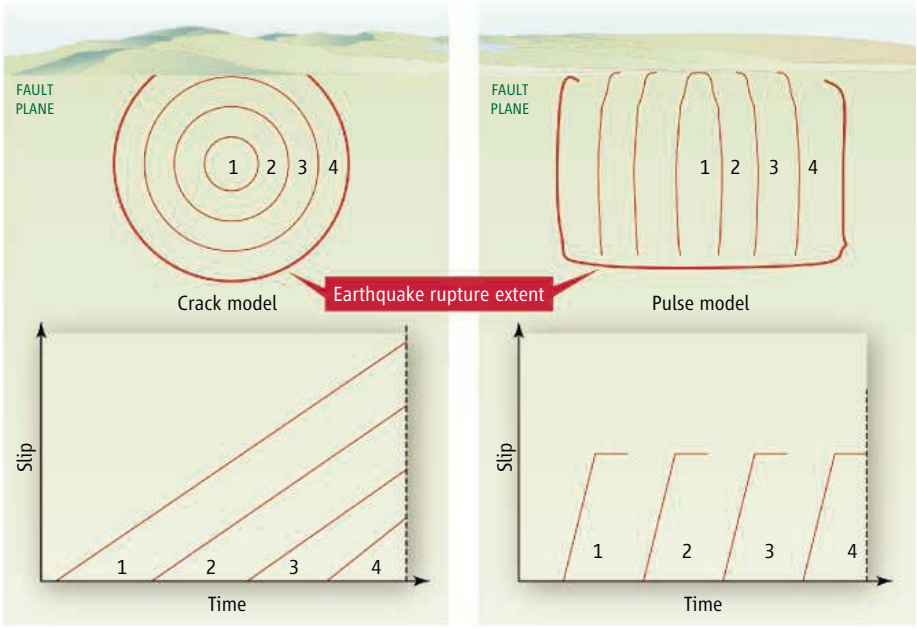
Violent shaking and destruction caused by earthquakes are the result of rupture and frictional slip on tectonic faults, and bigger earthquakes break bigger fault segments. But how do brittle ruptures of Earth's crust grow? Seismologic evidence shows that quakes begin in a small nucleation region and propagate at speeds up to 16,000 kilometers per hour. Two competing models of rupture growth describe this expansion (see the figure). In the crack model, the nucleation region slips throughout the quake and the slipping region expands until the rupture stops, a process akin to stretching a penny into the size of a half-dollar. In the pulse model, only a small portion of the total fault area slips at any one time, so as to cover the fault surface the way an inchworm crawls. Distinguishing between the two models is important for hazard assessment because they predict different degrees of strong shaking and ground acceleration with distance from the nucleation site. Recent seismological observations favor the pulse model, but efforts to connect these data with theoretical models of earthquake physics have been stymied because rupture pulses have never been reliably observed in the laboratory. However, as reported on page 1765 by Lykotrafitis *et al.* (*1*), new laboratory experimental evidence on brittle fracture, showing the existence of pulse-like ruptures and the conditions under which they exist, may help resolve the debate.

Lykotrafitis and co-workers sheared photoelastic material in frictional contact in a dynamic impact apparatus and monitored rupture propagation with high-speed photography. They show that the rupture propagation mode varies systematically with the strength of initial forcing (as produced by impact speed). Pulse-like ruptures are favored by slower impact speeds relative to those for crack-like ruptures. Also, the frictional slip velocity during rupture is lower for pulse-like ruptures than for crack-like ruptures. Thus, pulse-mode ruptures are the slow cousins of breaks that propagate as classical cracks. The data of Lykotrafitis *et al.* show a clear relationship between stress level and rupture propagation mode, with larger shear stress levels resulting in crack-like propagation.

The experiments of Lykotrafitis *et al.* address perhaps the most important question in earthquake physics: What controls seismic slip at a point on a fault? Virtually every quantifiable aspect of earthquakes depends on slip, but local fault slip cannot be measured directly from seismograms. If the initial tectonic shear stress determines slip, it would imply that dynamic frictional strength is zero and that stress on the fault drops to zero during an earthquake. In this scenario, seismic slip ceases because the local energy budget is depleted, but this runs counter to laboratory data on frictional stick-slip and seismic estimates of radiated energy, which indicate that seismic stress drop is a mere 10% of the tectonic stress level. Or, if the boundary conditions of fault strength determine seismic slip, then earthquake rupture stops when it encounters a strong barrier. Alternatively, frictional behavior during rupture—possibly abetted by dynamic variations in normal stress—could determine slip. The self-healing pulse model belongs to this last class of models. In order for rupture to propagate as a slip pulse, the fault must strengthen rapidly after slip so that local slippage ceases.

The crack model of earthquake rupture emerged in the 1970s as an extension of the mechanics of dislocations in solids, and much progress has been made in connecting seismic phenomena with the mechanics of

The authors are in the Department of Geosciences, Pennsylvania State University, University Park, PA 16802, USA. E-mail: cjm38@psu.edu



Rupture mechanisms. The diagram shows two different ways that ruptures might occur in a section of Earth (the fault plane faces front). In the crack model (**top left**), the nucleation region 1 slips throughout the quake, the slipping region expands until rupture stops, and the entire fault plane slips simultaneously. The lines (**bottom left**) represent slip history for four regions on the fault plane. In the pulse model (**top right**), only a small portion of the total fault area slips at any one time. All points on a pulse-mode rupture plane exhibit identical slip histories (**bottom right**).

dynamically propagating cracks. In the past decade, this model has been challenged by seismic observations and theoretical work that support a slip-pulse model. However, modern earthquake science (*2*) is based on the recognition that seismic records contain a spectral signature of rupture, with larger quakes releasing longer period waves such that the maximum period scales with rupture area. This observation dovetails nicely with crack models, for which the whole rupture area slips simultaneously, but presents a problem for self-healing pulses because the slipping region (see the figure), which represents the largest coherent spatial dimension, does not scale with overall rupture dimension. Moreover, earthquakes that propagate as pulses must accumulate the slip appropriate for the rupture size in a time window that itself does not scale with rupture duration.

There are several aspects of the scaling relationships among earthquake source parameters that are problematic for the pulse model. The simplest observable source parameters of earthquakes include rupture length and width, average slip, and the seismically induced reduction in local tectonic stress. A large catalog of data and their scaling relations are consistent with the crack model for earthquake rupture but require special (and in many cases physically implausible) circumstances to support the pulse model. For example, do ruptures know how big they will be when they begin to grow? Scaling relations indicate that small and large earthquakes begin the same way, but the pulse model does not easily fit within this framework. It is perhaps worth noting that these two end members are not the only possible rupture modes. In fact, the experiments discussed by Lykotrafitis *et al.* show both modes of rupture in the cases where pulses were generated. Extending these results by applying them to geologic materials and field observations will be the next challenge.

As permanent seismic detection networks increase in density and we accumulate high-quality seismic recordings close to large earthquakes, the mode of rupture propagation may be known routinely. But to understand the physical processes that govern dynamic rupture nucleation, growth, and arrest, we will need additional information provided by careful laboratory studies such as those described by Lykotrafitis *et al.*

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MOLECULAR BIOLOGY

Little Molecules with Big Goals

Bert W. O'Malley

Sets of master genes may control the expresson of the relevant proteins involved in complex cellular processes such as growth and metabolism.

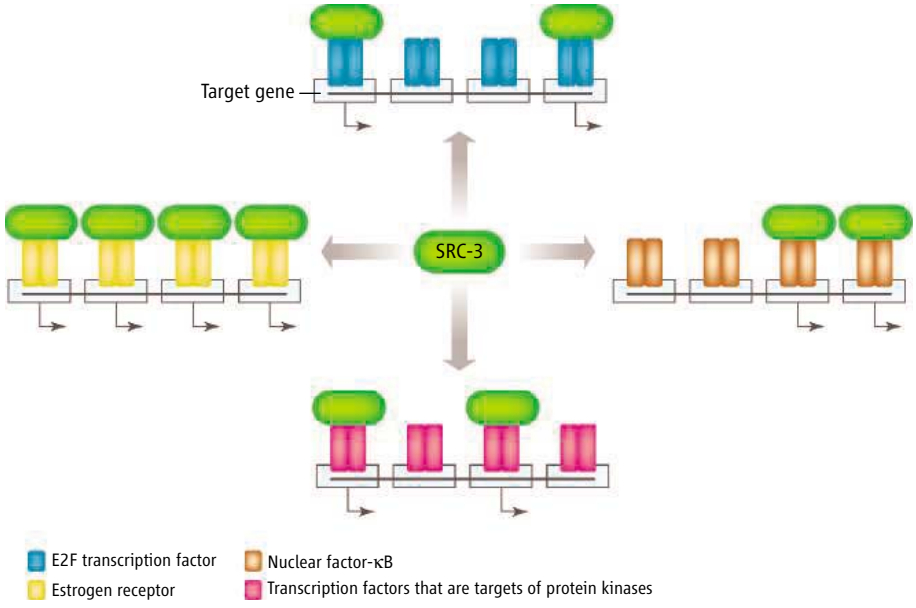
The field of nuclear receptor coregulators is approaching its 11th birthday (*1*), and the number of known constituents—coactivators and corepressors—has grown to more than 200 (*2*). Their characterization began modestly, with a simple concept that a limited number of coactivators existed that functioned merely as "adaptors" for stabilizing the cellular machinery that transcribes genes. But coregulator actions have expanded to chromatin modification and remodeling, initiation of transcription, RNA elongation and splicing, and protein degradation (*3*). In fact, we now know that coregulators comprise multiple (5 to 10) proteins of large regulatory machines, conveying the enzymatic activities needed to achieve diverse functions (*3*, *4*).

A central question emanates from the plethora of recent information on this class of regulatory molecules: Why have genes that code for so many coactivators and corepressors evolved? Are there big-picture goals behind their evolution, or do they exist simply to provide a series of disconnected catalytic reactions to activate or repress gene expression? Given the recent evidence for their expansive roles in biology, the latter seems clearly not to be the case. Rather, coregulators appear to constitute "little molecules with big goals" and likely represent the elusive "master genes" that were first proposed nearly 50 years ago (*5*), albeit different in form and substance.

The body controls, in a temporally and spatially coordinated manner, hundreds of genes needed to affect any single major complex process such as metabolism,

The author is in the Department of Molecular and Cellular Biology, Baylor College of Medicine, Houston, TX 77030, USA. E-mail: berto@bcm.edu

inflammation, and cell growth. It is now well accepted that multiple cooperating signaling pathways regulate such processes. But how can this be achieved when each signaling pathway depends on a battery of genomically dispersed genes that are activated by different DNA-binding transcription factors? Enter the coactivators, molecules that can move among hundreds of transcription factors and coordinately control the expression of appropriate subsets of genes to produce a desired end goal, such as growth. For example, a collection of coactivators including PGC-1, SRC-1, SRC-2, SRC-3, CBP/p300, and TRAP220 have achieved prominence in this regard (*6*), controlling numerous genes that regulate fat cell development and function, energy expenditure, and carbohydrate metabolism.



Multitasking molecules. Steroid receptor coactivator–3 (SRC-3) represents the product of a master gene, functioning as a coactivator for multiple subsets of genes that are under the control of different transcription factors in four diverse signaling pathways. This strategy accomplishes the overall functional goal of cell growth.

Another prime example of coordinated coregulator activity is during the even more complex processes of morphogenesis and growth. For example, in breast tissue, the gene encoding the coactivator SRC-3 exemplifies a "master gene," as it is implicated in coordinately regulating the expression of hundreds of genes involved in at least four different growth signaling pathways in this tissue (*7*) (see the figure).

Although our understanding of the molecular mechanisms of action of coactivators has rapidly progressed over the past decade (there is currently less known about corepressors), the clinical relevance of these molecules has garnered much attention in the etiology and progression of multiple pathologies. When the activities of the master genes are compromised, cellular processes can quickly deteriorate. For example, if a cancer cell is to achieve relentless growth, two major genetic choices are at hand. The oncogenic cell can activate many genes in multiple relevant signaling pathways to achieve simultaneous overproduction of proteins that will spur growth. On the other hand, it can simply overproduce a coactivator that then seeks out and coordinately controls the expression of all the requisite genes to allow the malignant cell to outstrip its peers in growth. Indeed, overexpression of the coactivator SRC-3 promotes oncogenic growth by just this mechanism, and SRC-3 is now accepted as an authentic oncogene in breast (*8*). In turn, the regulators, which function by posttranslational mechanisms to control the amounts and activities of coactivators, could then be considered to function as tumor suppressors.

This master gene concept could be applied repeatedly to complex disorders such as diabetes, inflammatory diseases, cardiovascular disease, as well as to other general physiological processes such as memory, learning, and reproductive functions. The picture that emerges is that for any single physiologic condition, a limited number of coregulatory molecules may control all the functionally relevant genes. This is precisely what Britten and Davidson had in mind when they first conceived the master gene concept, here modified so that "integrators" are coactivator proteins (rather than RNAs) and that the proposed "activator RNAs" are actually coactivator complexes.

Although there are some diseases that are clearly caused by defects in one gene, the single-gene concept is greatly overemphasized, and many disorders have complex multigenic etiologies. How could humans evolve multigene diseases with a similar phenotypic outcome but in genetically different families and ethnic groups? One way would be to inherit an array of multiple gene defects and then to acquire certain other complementary gene defects, all of which add up to an adult disease phenotype. However, there is a far easier means by which these multigenic disorders can occur. One could either inherit or acquire a defective allele (an alternate form of a gene) encoding one (or a few) coactivator(s) for the relevant multigenic pathway. By this mechanism, a coactivator (or corepressor) defect could simultaneously produce a coordinated misregulation of a subset of target genes in multiple different pathways. If you accept the published molecular evidence, then a multigene disorder could logically be the result. Ironically, the underlying cause of such a multigenic disorder could, in fact, be a single (or limited-number) gene malfunction.

As in all aspects of life, there is good news and bad news that can result from the evolution of a set of master coregulator genes in our genome. The good news is that the coregulator genes have provided us some of the molecular tools for making the jump in evolution that apparently has occurred in animals and humans, by permitting the coordinate regulation of diverse genetic activities required for complex functions. The bad news is that when things go awry, a corresponding rapid and coordinated development of a complex pathologic phenotype is a likely outcome. With the elucidation of these new potential drug targets, our charge for the future is to now translate the fundamental science of coregulators into the development of effective pharmaceuticals.

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SUMO1 Haploinsufficiency Leads to Cleft Lip and Palate

Fowzan S. Alkuraya,[1]* Irfan Saadi,[1]* Jennifer J. Lund,[1] Annick Turbe-Doan,[1] Cynthia C. Morton,[2] Richard L. Maas[1]†

Cleft lip with or without cleft palate (CL/P) is among the most common craniofacial birth defects. Several genes have been identified that contribute to CL/P, but the full spectrum of such genes and whether and how they interact is unknown. We identified a 5-year-old Caucasian girl born with unilateral cleft lip and palate (primary and secondary), who was otherwise phenotypically normal. Her karyotype was 46,XX,t(2;8)(q33.1;q24.3), and array CGH (comparative genomic hybridization) analysis was normal. Because the patient carried a balanced reciprocal translocation, we hypothesized that a gene important for palatogenesis was disrupted by the translocation. FISH (fluorescent in situ hybridization) analysis and suppression PCR (polymerase chain reaction) revealed that the *SUMO1* (small ubiquitin-related modifier) gene was interrupted by the 2q breakpoint, and *SUMO1* haploinsufficiency was further confirmed at the RNA and protein levels (Fig. 1A and fig. S1).

SUMO1 encodes a 101-residue polypeptide involved in posttranslational modification of many proteins (*1*). To establish a causative role for *SUMO1* haploinsufficiency in the pathogenesis of CL/P, we first examined the expression of murine *Sumo1* by whole-mount in situ hybridization at embryonic day 13.5 (E13.5) and observed strong expression in the upper lip, primary palate, and medial edge epithelia of the secondary palate (fig. S2). At E14.5, expression of *Sumo1* could be seen in the medial edge epithelial seam by using section in situ hybridization (fig. S2).

Next, we used an existing embryonic stem cell line in which *Sumo1* transcripts were interrupted by insertion of a β-*galactosidase*–expressing gene-trap vector into the *Sumo1* locus to generate $Sumo1^{Gt(pGT1Lxf)Byg}$ (henceforth referred to as $Sumo1^{Gt}$) mouse mutants. Wild-type transcripts were reduced in both hetero- and homozygotes, and variable reduction of Sumo1 protein was seen in heterozygotes. However, by X-gal staining, we detected that this hypomorphic allele faithfully recapitulated endogenous *Sumo1* expression in palatal shelf epithelium and mesenchyme at E13.5 (Fig. 1B). Several other genes required for palatogenesis, including *Eya1* (*2*) and *Msx1* (*3*), are expressed in palatal epithelium and/or mesenchyme at this time.

Among $Sumo1^{Gt/+}$ pups and embryos, 4 out of 46 (8.7%) exhibited cleft palate (Fig. 1C) or oblique facial cleft, compared with none in wild type ($n > 100$). In addition, the genotype distribution from heterozygous crosses at P1 (1:1.15:0.75) deviated from the expected 1:2:1. Both embryonic demise between E13.5 and E18.5 and immediate postnatal demise were noted (for *Sumo1* hetero- and homozygotes), indicating that *Sumo1* is required for other developmental functions besides palatogenesis.

Proteins encoded by three other genes, *MSX1*, *SATB2*, and *SMAD4*, are sumoylated and are either involved in or linked to pathways involved in palate morphogenesis (*4*–*7*). This suggests that *SUMO1* might control the activity of a repertoire of downstream effectors involved in palatogenesis, accounting for the sensitivity of palatal development to *SUMO1* gene dosage. To help place *SUMO1* in a molecular pathway relevant to its proposed role in CL/P, we tested for interaction between *SUMO1* and other cleft palate genes. Given its overlapping expression pattern with *Sumo1* in palatal shelf epithelium and mesenchyme, *Eya1* was an attractive candidate (fig. S2). Indeed, in $Sumo1^{Gt/+}$, $Eya1^{+/-}$ compound heterozygotes, the occurrence of cleft palate (36%) was significantly increased compared with that in $Sumo1^{Gt/+}$ (8.7%) ($P < 0.037$, Fisher exact test), or $Eya1^{+/-}$ (0%). Furthermore, we found Eya1 to be a substrate for sumoylation with SUMO1 in vivo. This was confirmed by abolishing the sumoylated Eya1 species with a SUMO-specific peptidase, SENP1. Furthermore, an Eya1 mutant protein in which two of three predicted high probability lysine residues were replaced with arginine displayed minimal sumoylation (Fig. 1D). These results identify a specific role for *SUMO1* in mammalian development and suggest that sumoylation regulates a network of genes that converge in palate development.

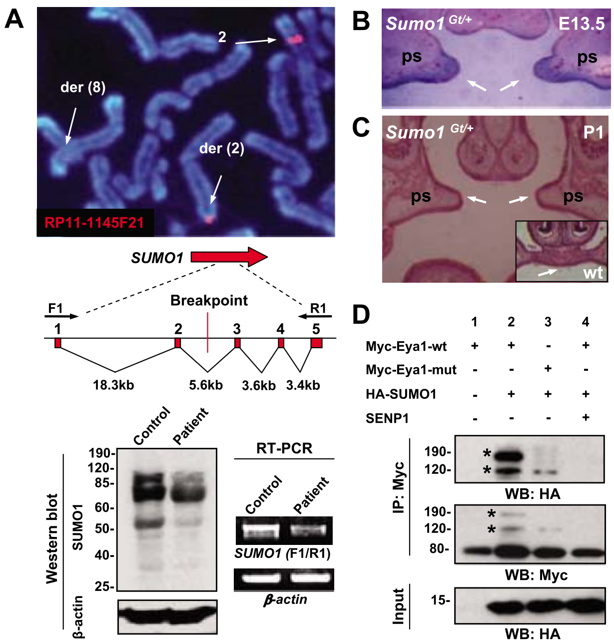

Fig. 1. (**A**) FISH using a clone that spans *SUMO1*. Location of the breakpoint is shown schematically. F1-R1 primers were used in reverse transcription PCR to show reduction of *SUMO1* expression in patient. Western blot shows reduced protein sumoylation in patient lymphoblasts compared with that of the control. (**B**) Coronal section of an E13.5 $Sumo1^{Gt/+}$ head showing *lacZ* expression in palatal shelf (ps) epithelium and mesenchyme. (**C**) Coronal section of a P1 $Sumo1^{Gt/+}$ head with ps elevated but not fused. (Inset) Normal control. (**D**) In the presence of hemagglutinin (HA)-tagged SUMO1, Myc-tagged Eya1 migrates as larger sumoylated species (*, lane 2), which disappear with the SUMO-specific peptidase, SENP1 (lane 4); the Eya1 Lys[43]→Arg[43]/Lys[459]→Arg[459] (K43R/K459R) (Eya1-mut) shows markedly reduced sumoylation (lane 3).

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Supporting Online Material

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[1]Division of Genetics, Department of Medicine, Brigham and Women's Hospital and Harvard Medical School, NRB-458, 77 Louis Pasteur, Boston, MA 02115, USA. [2]Department of Obstetrics and Gynecology and Reproductive Biology and Department of Pathology, Brigham and Women's Hospital and Harvard Medical School, NRB-160d, 77 Louis Pasteur, Boston, MA 02115, USA.

*These authors contributed equally to this work.
†To whom correspondence should be addressed. E-mail: maas@genetics.med.harvard.edu

Structural Basis of *glmS* Ribozyme Activation by Glucosamine-6-Phosphate

Daniel J. Klein and Adrian R. Ferré-D'Amaré*

The *glmS* ribozyme is the only natural catalytic RNA known to require a small-molecule activator for catalysis. This catalytic RNA functions as a riboswitch, with activator-dependent RNA cleavage regulating *glmS* messenger RNA expression. We report crystal structures of the *glmS* ribozyme in precleavage states that are unliganded or bound to the competitive inhibitor glucose-6-phosphate and in the postcleavage state. All structures superimpose closely, revealing a remarkably rigid RNA that contains a preformed active and coenzyme-binding site. Unlike other riboswitches, the *glmS* ribozyme binds its activator in an open, solvent-accessible pocket. Our structures suggest that the amine group of the *glmS* ribozyme-bound coenzyme performs general acid-base and electrostatic catalysis.

The *glmS* ribozyme is a catalytic RNA derived from the 5′-untranslated region (UTR) of the messenger RNA (mRNA) encoding glucosamine-6-phosphate synthase in numerous Gram-positive bacteria (*1*). It was identified computationally as a candidate riboswitch (*2*). Riboswitches regulate gene expression at the mRNA level, typically by undergoing structural rearrangements upon binding their cognate small molecules, which are often products of enzymes whose expression they regulate (*3*–*10*). Chemical probing of the *glmS* 5′-UTR produced little evidence of structural rearrangements upon binding of its cognate ligand, glucosamine-6-phosphate (GlcN6P) (*1*, *11*). Instead, the *glmS* 5′-UTR was found to undergo specific self-cleavage that is accelerated more than 10^5-fold by GlcN6P (*1*, *12*, *13*). In vivo analyses suggest that self-cleavage by this ribozyme regulates *glmS* gene expression and thereby synthesis of GlcN6P, a key metabolic precursor of the bacterial cell wall (*1*). The *glmS* ribozyme is the first example of a catalytic riboswitch.

The *glmS* ribozyme is also the first self-cleaving RNA identified in the mRNA of free-living organisms. Uniquely among natural ribozymes, its activity is modulated by the binding of a small molecule metabolite. However, like the four other natural self-cleaving RNAs (*14*), its cleavage reaction is a transesterification that produces 5′-OH and 2′,3′-cyclic phosphate termini, and it can be engineered into a multiple-turnover catalyst that cleaves substrate RNAs in trans (*2*). Studies of these four ribozymes have shown that their folds and active site structures are distinctly different and that they use a variety of sophisticated catalytic strategies (*15*–*18*).

In their original report, Winkler *et al*. did not establish whether GlcN6P participates directly in catalysis or activates the *glmS* ribozyme by allosterically inducing a conformational change (*1*). Recent biochemical data are consistent with the former (*11*, *12*, *19*). A variety of small molecules, which have in common with GlcN6P a hydroxyl group located β to an amine, can also activate *glmS* ribozyme cleavage in vitro, albeit to a much lesser extent (*12*). However, glucose-6-phosphate (Glc6P), which is isosteric with GlcN6P but lacks the amine group, is a competitive inhibitor (i.e., an antagonist) (*12*). These results are consistent with a coenzyme function for GlcN6P.

We report structures of a full-length *glmS* ribozyme bound to the competitive inhibitor Glc6P, as well as in the unliganded and product states. Comparison of the three structures demonstrates that the RNA is highly rigid and contains a solvent-accessible coenzyme binding pocket. Our ribozyme catalyzes rapid, GlcN6P-dependent cleavage in the crystalline state, further indicating that activation and catalysis proceed without substantial RNA rearrangements. Our structures indicate how GlcN6P is specifically recognized, how water molecules and a divalent cation mediate RNA-activator interaction, and how the amine functional group of GlcN6P is precisely positioned to serve as a coenzyme within a preorganized active site.

Structure determinations. Previously, it was shown that truncation of the *Bacillus subtilis glmS* 5′-UTR to 1 nucleotide (nt) 5′ and ~150 nt 3′ of the scissile phosphate, or deletion of the P1 loop, resulted in RNAs fully active in vitro (*1*, *2*). Deletion of the P1 loop yields *glmS* RNAs composed of two strands: a substrate oligonucleotide and a longer ribozyme. We crystallized two-strand constructs of the *glmS* ribozyme from *Thermoanaerobacter tengcongensis* (*20*). The postcleavage state ribozyme structure was solved at 2.1 Å resolution by multiple isomorphous replacement. Replacement of the 2′OH nucleophile of the substrate with other functional groups has been previously used to trap ribozymes in precleavage states (*14*). We determined the structure of a similarly trapped precleavage state *glmS* ribozyme bound to the competitive inhibitor Glc6P at 2.7 Å resolution. We also solved structures of two different trapped precleavage state RNAs by using crystals grown and stabilized in either the presence or the absence of GlcN6P at resolutions between 2.1 and 2.35 Å. Although binding of GlcN6P was undetectable crystallographically in these structures, we found that addition of GlcN6P to crystals of our *glmS* ribozyme containing a cleavable substrate resulted in specific cleavage in the crystal (*21*).

Overall structure of the *glmS* ribozyme. Three coaxial stacks of RNA helices packed side by side are the dominant feature of *glmS* ribozyme tertiary structure (Fig. 1). The stack encompassed by P1 and P3.1 is the longest (~100 Å). This stack and a smaller stack composed of P4 and P4.1 sandwich a short central stack, P2.1. Near-parallel packing of the three stacks results in a molecule that is ~50 Å wide but only ~20 Å deep. Helices previously proposed on the basis of covariation analyses are observed in our crystal structures, except P2a. Nucleotides predicted to form P2a instead contribute to helices P2.1 and P2.2 (Fig. 1C and fig. S1).

A doubly pseudoknotted ribozyme core. Previous analyses demonstrated that truncated *glmS* RNAs spanning from nucleotide –1 to the end of helix P2 retain a reduced amount of GlcN6P-dependent catalytic activity (*1*, *13*). Our structures reveal that this minimal sequence folds into a compact, contiguous arrangement that comprises both the active site and the metabolite-binding pocket. Indeed, the active site on the front of the molecule (Fig. 1A) and the metabolite-binding pocket on the back (Fig. 1B) are separated only by the scissile phosphate. This ribozyme core consists of a double pseudoknot that positions the central helix P2.1 with its major groove cradling the scissile phosphate. The adjacent helix P2.2 bears the scissile phosphate at its 5′ end, and its major groove is engaged in metabolite binding. Precise positioning of the P2.1 and P2.2 double pseudoknot is accomplished by four nonhelical crossovers (red lines, Fig. 1C) that connect the P1-P2.2-P2 stack with P2.1, as well as by a tightly bound metal ion (Fig. 1A).

Nucleotides in the two upper crossovers, which form the roof of the active site, participate in conserved base triples (Fig. 2A) that brace the three-way junction between P1, P2.2, and P2.1. G^{34} occupies the major groove of P2.2 where it makes a base triple with G^7 and C^{60}, while the

Division of Basic Sciences, Fred Hutchinson Cancer Research Center, 1100 Fairview Avenue North, Seattle, WA 98109–1024, USA.

*To whom correspondence should be addressed. E-mail: aferre@fhcrc.org

next residue (A^{35}) occupies the major groove of P2.1, where it forms a base triple with C^{36} and G^{53}. Thus, the consecutive nucleotides G^{34}, A^{35}, and C^{36} connect the P2.2 and P2.1 stacks. The conserved A^{54} and U^{59} make an unusual trans Watson-Crick base pair, which stacks on the A^{35}-C^{36}-G^{53} platform. The floor of the active site results from threading G^{66} and U^{67} through the closed loop between P2 and P2.1 (Fig. 2B). G^{66} and U^{67} are splayed apart so that their nucleobases stack on nucleotides of either of the lower crossovers (yellow and purple, Fig. 2B). The lack of base-specific contacts accounts for the lesser conservation of G^{41}, G^{66}, and U^{67}.

A peripheral domain buttresses the core. *GlmS* RNA nucleotides 3′ to the ribozyme core constitute a peripheral domain comprising four helical segments: P3, P3.1, P4, and P4.1 (Fig. 1). As predicted (*13*, *22*), P3 and P3.1 stack coaxially and form a classical pseudoknot resembling those of plant viral genomic RNAs (*23*, *24*). P4 and P4.1 form a coaxial stack that buttresses P2.1 (Fig. 1). Among the interactions that position the peripheral domain are the coaxial stacking of P2 and P3 and a class I A-minor interaction (*25*) between A^{117} of the P4.1 tetraloop and C^{10}-G^{31} of P1. In addition, a purine stack formed by the internal loop between P4 and P4.1 directly buttresses the minor groove of P2.1 (Fig. 2C). This interface differs from a stack of canonical A-minor motifs because of the oblique angle (~70°) between the axes of the purine stack and the P2.1 helix. Whereas adenosines in canonical A-minor motifs contact a single base pair (*25*), the high obliquity of the helical axes in the *glmS* ribozyme allows A^{105} and G^{128} to contact two, and A^{104} and A^{127} to contact three, consecutive base pairs (Fig. 2C), resulting in the most solvent-inaccessible interface in the ribozyme structure. Association of the core and peripheral domains buries ~2500 Å^2 of solvent-accessible surface area.

A rigid active site devoid of metal ions. We determined structures of the *glmS* ribozyme trapped in a precleavage conformation by replacing $A^{(-1)}$ with 2′-aminoadenosine or 2′-deoxyadenosine. The structures of these precleavage state complexes are in excellent agreement (fig. S2). The active site is com-

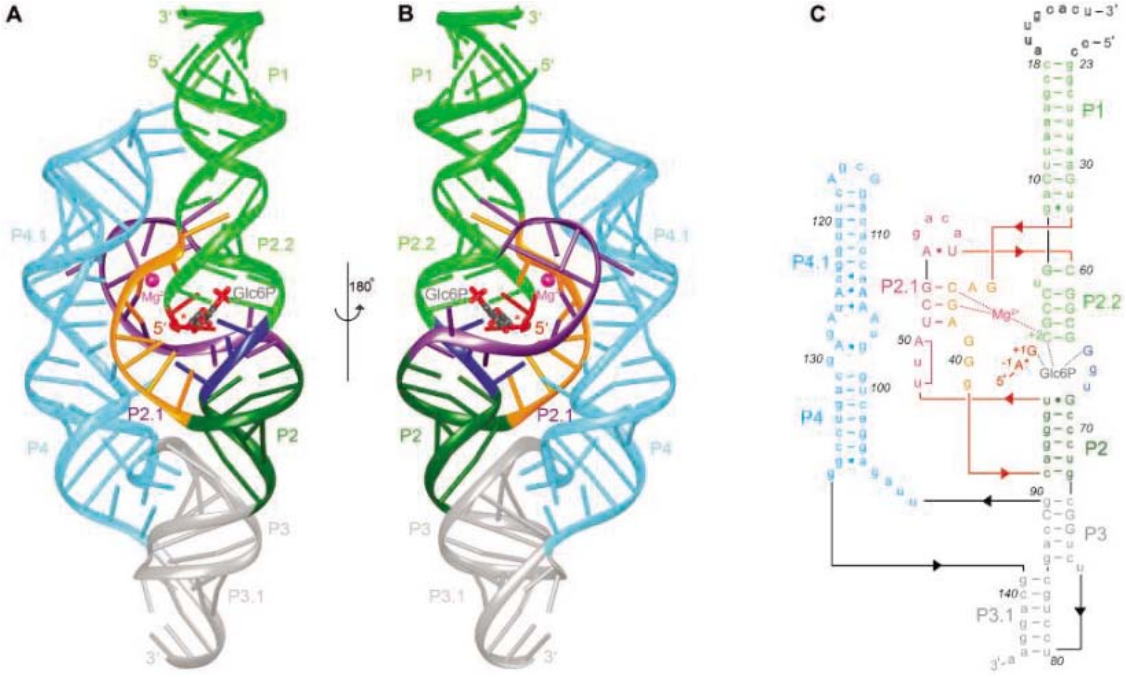

Fig. 1. Overall structure of the *glmS* ribozyme. (**A**) Cartoon representation of the Glc6P-bound state. The RNA chain 5′ to the scissile phosphate (*) enters the ribozyme from this side. The only crystallographically observed Mg^{2+} ion that makes multiple inner-sphere coordinations to the RNA is shown as a pink sphere. (**B**) View rotated 180°. Bound Glc6P is colored gray and red. (**C**) Revised secondary structure. Base-paired segments are named as in the literature (*1*, *22*), except P2.1 and P2.2, which were not predicted. Uppercase letters indicate nucleotides that are >90% conserved among known sequences (*13*). P1 nucleotides in black outline were engineered for crystallization and are not from the natural RNA.

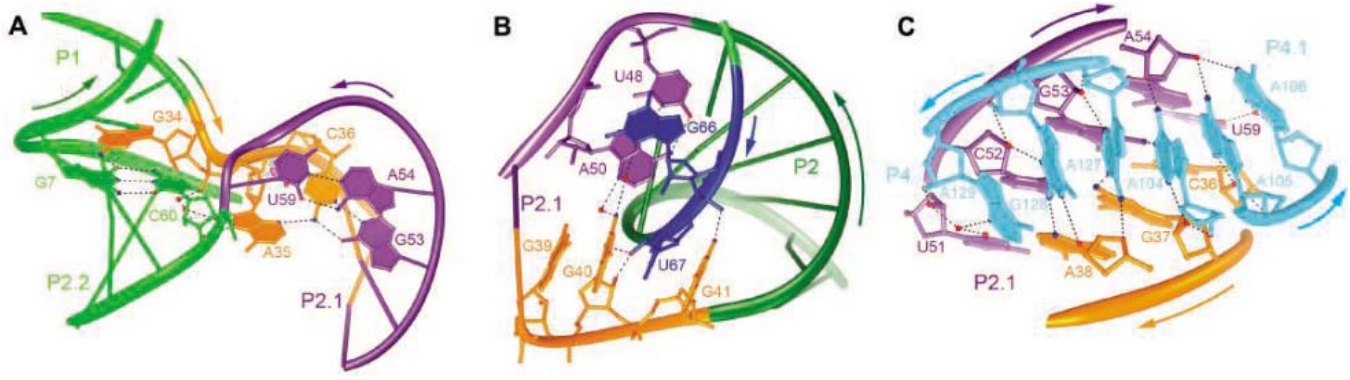

Fig. 2. Conserved tertiary interactions that shape the *glmS* ribozyme. Colors and numbering in all figures follow Fig. 1, except where indicated. Dashed lines depict hydrogen bonds. (**A**) Three-way junction between P1, P2.1, and P2.2, showing side-by-side base triples (view approximately as in Fig. 1B). A^{35} and the major grooves of P2.1 and P2.2 form the roof of the active site and the metabolite-binding pocket. (**B**) Floor of the active site, formed by threading G^{66} and U^{67} through the closed loop between P2 and P2.1. View is downward from the scissile phosphate (compare with Fig. 1A). (**C**) Oblique purine stack at the interface between P4 and P2.1.

posed mostly of nucleotides that are absolutely conserved across phylogeny (fig. S3) and lacks metal ions positioned for catalysis. The closest crystallographically observed metal ion is ~10 Å from the scissile phosphate and plays a structural role. $A^{(-1)}$ and G^{1} occupy a channel lined by the major groove of P2.1 and the bases of G^{39}, G^{40}, G^{65}, and G^{66} (Fig. 3A). The conserved G^{1} stacks beneath A^{35} but makes no base-specific contacts in the unliganded structures. The invariant $A^{(-1)}$ makes a specific trans sugar edge base pair with G^{65}, which explains the deleterious effect of G^{65} mutants on ribozyme activity (*26*). G^{39} and G^{65} position the scissile phosphate by hydrogen bonding to either of its nonbridging oxygens. Together, these interactions and the 2′-endo ribose pucker of $A^{(-1)}$ twist the RNA backbone into a conformation very close to that required for in-line attack of the scissile phosphate by the 2′-OH of $A^{(-1)}$. The angle between nucleophile, electrophile, and leaving group (τ) is 155° in our 2′-aminoadenosine *glmS* precleavage structure. This is close to ideal alignment [180° (*27*)], and comparable to the 158° τ angle observed in precleavage hairpin ribozyme structures (*28*, *29*).

To identify conformational changes that accompany *glmS* ribozyme activity, we determined the postcleavage state structure by using an oligonucleotide beginning with G^{1}. In the active site, G^{1} remains stacked beneath A^{35} (Fig. 3B). However, its free 5′-OH swings around to hydrogen bond with N6 of A^{38}. Subtle rotations of G^{39} and G^{65} are also observed, which may be due to the absence of $A^{(-1)}$ and the scissile phosphate to which they hydrogen bond in the precleavage state (Fig. 3A). Except for these small differences, the pre- and postcleavage structures superimpose very closely (Fig. 3C) with an all-atom rmsd of 0.51 Å, comparable to the mean precision of our structures (table S1).

Metabolite-binding pocket. By using crystals of the precleavage state ribozyme assembled with our 2′-deoxy RNA inhibitor, we discovered that the competitive inhibitor Glc6P (*12*), which differs from GlcN6P by a single atom, binds in a conserved pocket behind the active site (Fig. 4, A and B, and fig. S3B). The unbiased experimental electron density (Fig. 4C) unambiguously indicates that the metabolite binds as the α-anomer. The *glmS* ribozyme recognizes both sugar and phosphate moieties of Glc6P (Fig. 4C), burying ~80% of the solvent-accessible surface of the metabolite. The unpaired nucleobase of G^{1} stacks on the glucopyranoside ring while simultaneously donating a hydrogen bond from its N1 to the phosphate of Glc6P. The phosphate of Glc6P also coordinates a Mg^{2+} ion, which in turn coordinates O6 of G^{64}. The anomeric hydroxyl of Glc6P donates a hydrogen bond to the pro-*R*p oxygen of the scissile phosphate and receives a hydrogen bond from N1 of G^{65}. The 2-OH of Glc6P hydrogen bonds to the 5′ oxygen of G^{1}, as well as a tightly bound water molecule, and is within ~3.6 Å of the scissile phosphate. The 3-OH of Glc6P hydrogen bonds to the 2′-OH of A^{50}. The ribozyme makes no direct interactions with the 4-OH of Glc6P. O5 of Glc6P receives a hydrogen bond from N4 of C^{2}.

The *glmS* ribozyme is active in the crystalline state. The apparent inability of our trapped precleavage state *glmS* ribozyme crystals to bind GlcN6P could indicate that crystallization has trapped the ribozyme in an inactive conformation. To address this possibility, we crystallized *glmS* ribozymes assembled with an all-RNA substrate in the absence of both Glc6P and GlcN6P. The low GlcN6P-independent cleavage rate (*12*) suggested that the substrate would remain intact during the 5 days required for crystallization. Two crystals obtained in this manner were soaked for 5 min in either Glc6P or GlcN6P, flash frozen, and used for data collection. The first crystal yielded unambiguous electron density for $A^{(-1)}$ as well as Glc6P (Fig. 5A)

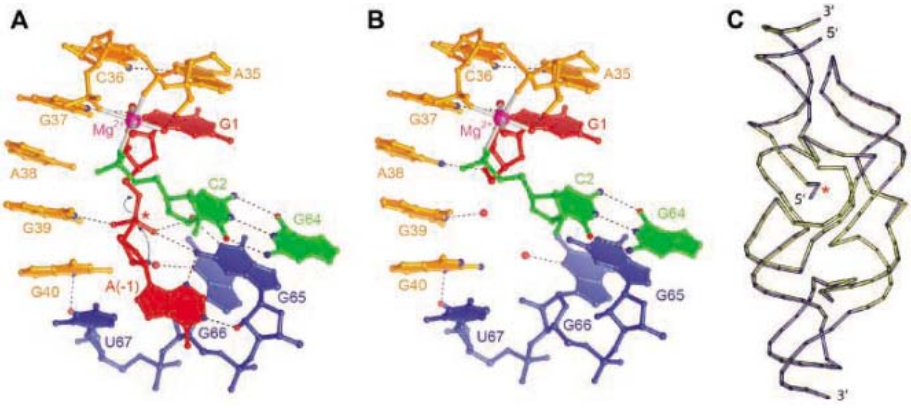

Fig. 3. A rigid, preorganized active site. (**A**) 2′-aminoadenosine inhibitor-bound precleavage active site highlighting interactions that position reactive groups for catalysis (view approximately as in Fig. 1A). Gray cylinders depict coordination to the tightly bound magnesium ion. Free-floating red spheres are ordered water molecules. (**B**) Structure of the postcleavage state. (**C**) Backbone superposition of the precleavage (blue) and postcleavage (yellow) state structures demonstrating the absence of substantial conformational changes between the two states (view as in Fig. 1B). Structures were superimposed with the use of all atoms between G^{1} and A^{145}, inclusive.

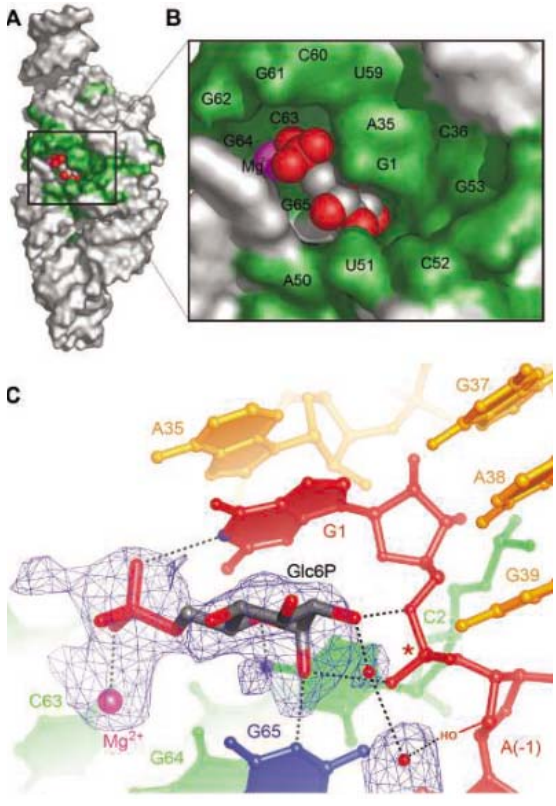

Fig. 4. Coenzyme binding pocket. (**A**) Molecular surface of the *glmS* ribozyme (orientation corresponds to Fig. 1B). Nucleotides >90% conserved are colored green. Atomic spheres are shown for Glc6P and the Mg^{2+} ion that coordinates its phosphate. (**B**) Expanded view of the Glc6P binding pocket. (**C**) Portion of the Glc6P-bound, 2′-deoxy $A^{(-1)}$ precleavage structure superimposed on the residual simulated-annealing omit $|F_o| - |F_c|$ electron density map contoured at 2.5σ [orientation is rotated slightly from (A)] and a stick figure of the bound Glc6P. Hydrogen bonding interactions involving Glc6P and two active site water molecules (red spheres) are shown. The position of a modeled 2′-OH of $A^{(-1)}$ is shown to indicate its proximity to one of the two buried waters.

and resulted in a structure indistinguishable from that obtained with the 2′-deoxy-RNA inhibitor bound with Glc6P, except for presence of the 2′-OH of $A^{(-1)}$. The second crystal yielded a map that lacked electron density for $A^{(-1)}$ as well as GlcN6P (Fig. 5B) and resulted in a structure indistinguishable from our previous postcleavage structure. This demonstrates that rapid, GlcN6P-dependent RNA cleavage is catalyzed by the *glmS* ribozyme in the crystalline state. Furthermore, the 5′ cleavage product and GlcN6P dissociate from the active site after the reaction. Together, these results argue that the structures we have solved depict the *glmS* ribozyme in an activator-responsive conformation. This suggests that the chemical identity of the 2′-functional group of $A^{(-1)}$ indirectly affects GlcN6P (but not Glc6P) binding. The ribosome (*30*) is the only other ribozyme for which activity has been visualized in crystals.

Catalytic mechanism and discussion. The specific interactions that we observe between the *glmS* ribozyme and Glc6P fully explain the previously determined activities of various compounds (*12*) if the 2-amine of GlcN6P were to occupy the same position as the 2-OH of Glc6P. That glucosamine is a weaker activator than GlcN6P (*12*) is consistent with its inability to interact with the N1 of G^1 through a phosphate. The ability of various β-hydroxylamines (such as ethanolamine, serinol, and tris) to activate the *glmS* ribozyme (*12*) is consistent with their capacity to present their hydroxyl and amine groups in the same relative orientation as the anomeric- and 2-OH groups, respectively, of Glc6P. The modest amount of activation achieved by these simpler amines presumably reflects their inability to make additional interactions with the RNA. Most compelling is the activation by L-serine but not D-serine (*12*). Only L-serine has the chirality to orient its γ oxygen and amine in the same way as the crystallographically observed anomeric- and 2-OH, respectively, of Glc6P. The explanatory power of our structure, the isostery between Glc6P and GlcN6P, and the kinetic competition between Glc6P and GlcN6P (*12*) strongly argue that the crystallographically determined mode of binding of Glc6P depicts the catalytically productive mode of binding of GlcN6P.

Binding of GlcN6P in the exact location occupied by Glc6P would allow the activator to achieve catalysis by as many as three means. First, the amine of GlcN6P could serve as a general base. Although the 2′-OH nucleophile of $A^{(-1)}$ is sterically inaccessible to GlcN6P, our structures reveal a buried water molecule that hydrogen bonds to the nucleophile and a second water molecule that would bridge the amine of the activator with the first buried water (Fig. 4C). Thus, the metabolite may activate the nucleophile indirectly through a proton relay (Fig. 6A). Alternatively, the N1 of G^{40}, which is located 3.2 Å from the nucleophile, could serve as a general base. Second, once the amine becomes positively charged, it is ideally placed to stabilize the increased negative charge of the pentacoordinate phosphorus transition state (Fig. 6B). Third, the ammonium ion resulting from protonation of GlcN6P could also serve as a general acid, donating a proton to the 5′-oxo leaving group to which it hydrogen bonds in the precleavage state (Fig. 4C).

Our structures demonstrate that the *glmS* ribozyme adopts a rigid fold with a doubly pseudoknotted core. This represents the second example [the first being the hepatitis delta virus ribozyme (*31*, *32*)] of a doubly pseudoknotted core in a natural self-cleaving ribozyme. Although the cores of the *glmS* and HDV ribozymes have some superficial three-dimensional similarity (fig. S4), the relative arrangement of major and minor grooves and the nucleobases surrounding the cleavage sites are completely different between these two catalytic RNAs. Thus, the *glmS* ribozyme is an independent evolutionary solution for catalyzing sequence-specific RNA self-cleavage.

The hairpin and the *glmS* ribozymes both use the energy of substrate strand binding to distort the nucleotides flanking the scissile phosphate into a reactive conformation that is held in a rigid active site. Unlike the hairpin ribozyme, which uses proximity effects (*28*), preferential transition-state binding (*18*), and possibly general acid-base catalysis by nucleobases (*33*–*36*), the *glmS* ribozyme is completely dependent on a coenzyme to carry out general acid-base and electrostatic catalysis. Our structures are consistent with the view that the *glmS* ribozyme has evolved to provide a rigid binding site for the substrate and its coenzyme (fig. S5), and has delegated (*1*, *12*, *37*) catalytic chemistry to the coenzyme. Indeed, it appears that the active site has a propensity for binding small

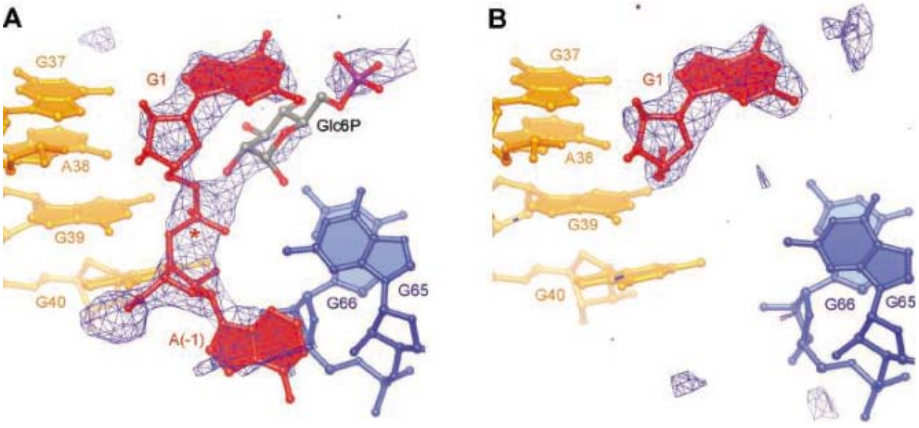

Fig. 5. GlcN6P-dependent cleavage of *glmS* ribozymes in the crystalline state. (**A**) Simulated-annealing omit $|F_o| - |F_c|$ electron density (contoured at 3.0σ) calculated with data from an all-RNA crystal soaked in Glc6P and phases from a model lacking $A^{(-1)}$, G^1, and Glc6P. (**B**) Simulated-annealing omit $|F_o| - |F_c|$ electron density (contoured at 3.0σ) calculated with data from an all-RNA crystal soaked in GlcN6P and phases from a model lacking $A^{(-1)}$, G^1, and Glc6P.

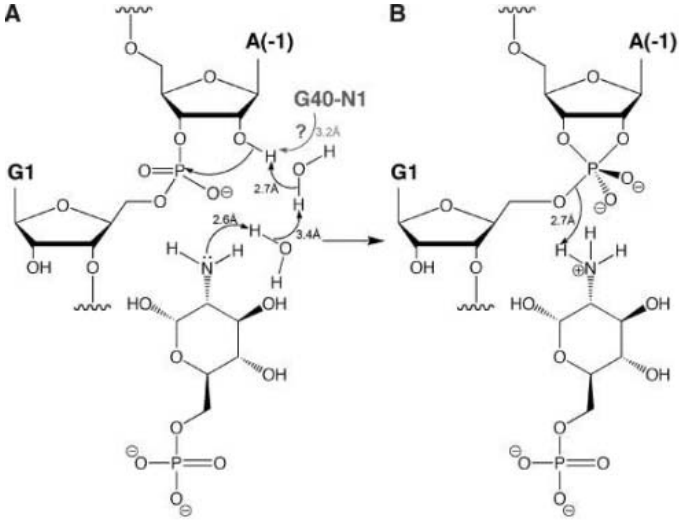

Fig. 6. Putative catalytic mechanism deduced from the Glc6P-bound crystal structure of the *glmS* ribozyme. (**A**) The 2-amine of GlcN6P may serve as a general base that deprotonates the 2′-OH nucleophile of the transesterification through two tightly bound water molecules. An alternative candidate for a general base that deprotonates the 2′-OH is the universally conserved G^{40}. (**B**) Electrostatic stabilization of the negatively charged pentacoordinate phosphorus transition state and protonation of the 5′-oxo leaving group by the 2-ammonium group of GlcN6P.

molecules that persists into the postcleavage state (fig. S6).

Conclusion. The *glmS* ribozyme differs from typical riboswitches in two fundamental ways. First, the *glmS* ribozyme adopts a rigid fold that does not change upon metabolite binding (fig. S5) or during the course of the reaction (Fig. 3C). In contrast, conventional riboswitches couple the energy derived from metabolite binding to substantial conformational change of the RNA and only adopt a stable fold once they bind their cognate metabolites (*3*, *4*, *6*, *7*, *9*, *10*). Second, unlike the purine, S-adenosylmethionine, and thiamine pyrophosphate riboswitches, which achieve specificity by completely encapsulating their ligands (*3*, *4*, *7*, *9*, *10*), the *glmS* ribozyme achieves high specificity and satisfies its requirement for a precisely positioned functional group with a pK_a suitable for general acid-base catalysis using a solvent-accessible binding pocket. The open and rigid coenzyme-binding pocket of the *glmS* ribozyme, which is only partially filled by the natural ligand (Fig. 4B), appears to be an excellent target for the development of novel small-molecule activators or inhibitors that may have antibiotic properties.

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Supporting Online Material

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Materials and Methods
SOM Text
Figs. S1 to S8
Tables S1 and S2
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REPORTS

Irreversible Organic Crystalline Chemistry Monitored in Real Time

Peter R. Poulin and Keith A. Nelson*

Because multiple laser shots are typically required to monitor ultrafast photochemical reaction dynamics, sample depletion and product accumulation have greatly restricted the range of substrates and structural environments amenable to study. By implementing a two-dimensional spatial delay gradient across the profile of a femtosecond probe pulse, we can monitor in a single laser shot organic crystalline reaction dynamics despite the formation of permanent photoproducts that cannot be conveniently removed. We monitored the photolysis of the triiodide anion, I_3^-, and subsequent recombination or relaxation of its reaction products, in three very different pure organic molecular crystals. The experimental results and associated molecular dynamics simulations illustrate the intimate connection between lattice structure and reaction dynamics, highlighting the role of lattice constraints in directing phase-coherent geminate recombination of photofragments within a crystalline reaction cage.

Direct time-resolved observation of chemistry in the solid state offers an experimental precision especially well suited to detailed modeling of local influences on reactivity. Advantages include the uniformity of the crystal lattice structure and the constraints it places on the motions of reactants and products; the limited number of well-defined lattice vibrational modes through which dynamical exchange of energy between the reacting species and the surroundings may occur; and the systematic variation of the lattice environment encountered in related members of a crystalline family (*1*–*4*). This potential for detailed analysis and understanding cannot be matched in liquid-state photochemistry, because even though short-time reaction dynamics in liquids also are strongly influenced by local intermolecular geometries and interactions, experimental measurements are invariably averaged over myriad local environments; consequently, the distinct time-dependent signatures of different surroundings are obscured. Mechanistic insight into the influence of crystal structure and local intermolecular topology on reaction dynamics and yields has been termed topochemistry (*1*) and discussed qualita-

Department of Chemistry, Massachusetts Institute of Technology, 77 Massachusetts Avenue, Cambridge, MA 02139, USA.

*To whom correspondence should be addressed. E-mail: kanelson@mit.edu

tively or semiquantitatively in terms of a reaction cage formed by the local lattice structure (*5*). Beyond its fundamental interest, such insight can guide the design of molecular crystals with applications ranging from solid-state organic synthesis to organic electronics to energetic materials. Direct time-resolved observation of ultrafast reaction dynamics in zeolites (*6*) and related porous structures would also be of considerable interest for catalyst optimization.

Unfortunately, the probing of ultrafast crystalline photochemistry using conventional techniques of femtosecond spectroscopy (*7*) is generally frustrated by the buildup of permanent reaction products and by the difficulty of replacing a degraded sample with fresh material, as achieved through flow in studies of liquid- or gas-phase samples. Thus, ultrafast spectroscopy of crystalline chemical reactions has been rare and has focused on a small set of reversible reactions in organic crystals and rare gas matrices (*8*). A generally applicable method for unrestricted study of photochemistry in complex organic lattice environments, including covalent bond breakage, molecular fragmentation into new chemical products, and product evolution, has been lacking. Here we apply a single-shot probing approach that reveals crystalline reaction dynamics with femtosecond resolution.

To gauge the impact of lattice environment on reactivity, we studied three pure molecular crystals, each containing the same photolabile anion, triiodide (I_3^-), and a different complex organic counterion (*9*). The substantial differences in lattice structure are evident from the crystal structures (Fig. 1) of tetra-*n*-butylammonium triiodide (TBAT) (*10*), tetraethylammonium triiodide (TEAT) (*11*), and tetraphenylphosphonium triiodide (TPPT), determined through x-ray diffraction.

Gas and liquid-state spectroscopy (*12*–*19*) have established that photoexcitation of I_3^- at an ultraviolet (UV) wavelength of 300 nm leads to rapid dissociation, yielding a bound I_2^- anion and a neutral I atom. The I_2^- species may be probed through its broad absorption band in the visible and near-infrared (IR) spectral regions without interference from the I_3^- UV absorption. In all three crystalline systems, exposure to a single weak UV excitation pulse (*9*) produces a clearly visible, permanent discoloration of the crystal at the site of irradiation. The usual approach to femtosecond spectroscopy of repeated sample excitation, with data accumulated over many thousands of laser shots, was clearly inapplicable to solution-grown crystals, each offering only a small region of acceptable optical quality for measurement.

To circumvent this problem, we developed a technique to acquire multiple time-resolved data points with a single laser pulse. The use of an echelon structure (Fig. 2B) to subdivide the transverse profile of a probe beam into multiple distinct pulses, each of which reaches the sample at a different time, was introduced in the early days of picosecond spectroscopy (*20*). However, the scheme was difficult to implement and was supplanted by the use of a variable delay line (Fig. 2A). The echelon

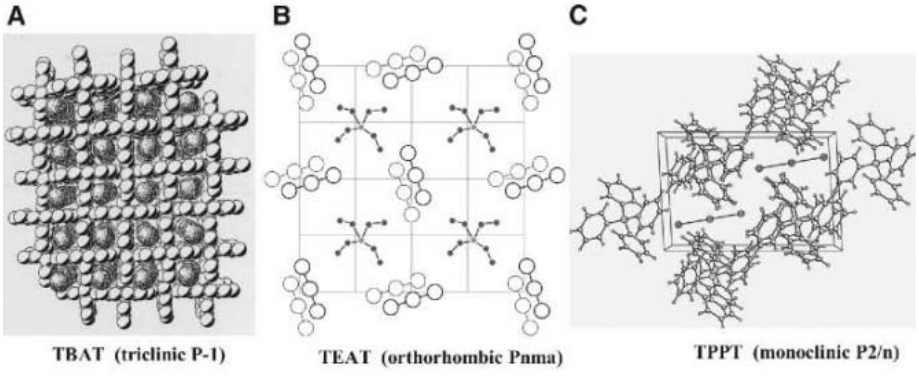


Fig. 1. Crystal structures of (**A**) tetra-*n*-butylammonium triiodide (TBAT), (**B**) tetraethylammonium triiodide (TEAT), and (**C**) tetraphenylphosphonium triiodide (TPPT). The structures are determined by x-ray diffraction. I_3^- ions are oriented in approximately longitudinal columnar chains in TBAT and in nearly orthogonal side-on stacks in TEAT. In TPPT, each I_3^- ion is situated in a local lattice pocket. The structures of TBAT and TPPT are derived from our x-ray data, and the depiction of TEAT is adapted from (*3*).

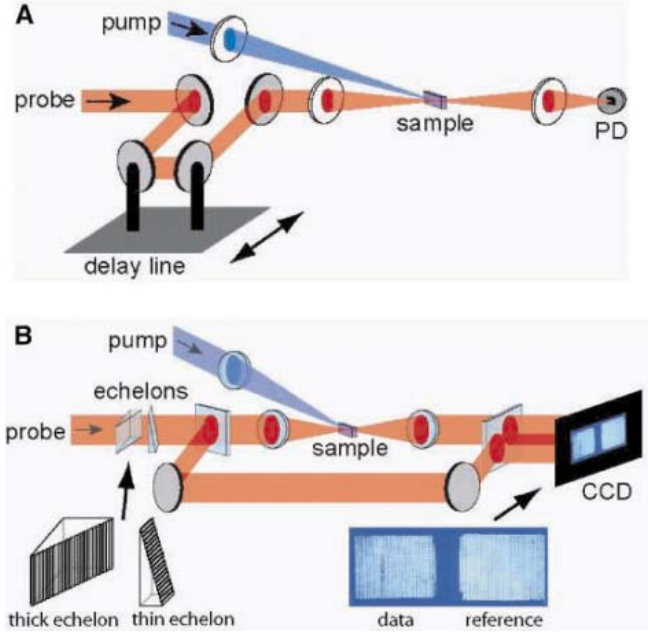


Fig. 2. Conventional and single-shot femtosecond spectroscopy. (**A**) In conventional femtosecond time-resolved measurements, an excitation (or pump) pulse initiates a reaction and a probe pulse monitors the evolution at a single, selected time delay *t* (PD, photodetector). The procedure is repeated for many different excitation-probe delays to provide a complete time-dependent data set. (**B**) In the present study, a complete data set is recorded following a single pump laser shot with 400 probe pulses that arrive at the sample successively delayed by 25-fs time increments and covering a total temporal range of 10 ps. The 400 pulses are formed from one original probe beam that is expanded in diameter and passed through two orthogonally oriented stair-step echelon structures. Portions of the probe pulse that pass through thicker combined glass regions are delayed by longer times. Each echelon has 20 steps, with step heights yielding relative delays of 25 fs in one structure and 500 fs in the other. The resulting 20 × 20 array of pulses is focused to a common spot at the sample position and then imaged onto a CCD camera, along with a reference image that bypasses the sample. The reference image is used to correct for variation in the incident beam spatial profile, imperfections in the echelon structures, and scattering of probe light due to imperfect sample optical quality. The CCD image shows data and reference images recorded from a single-shot measurement on a glass (fused silica) test sample. One vertical region of the data image shows substantial changes in probe pulse transmission through the sample, which are induced by the arrival of the excitation pulse. These changes last for ~300 fs (~12 distinct probe regions on the CCD image). The photoinduced responses of chemical interest in the present work are far smaller in magnitude and cannot be discerned by eye from the raw data images. The overall time resolution of the single-shot experimental setup, which is limited by probe pulse broadening within the echelons, the noncollinear pump-probe angles, and the optical properties of the samples, is ~50 fs.

approach is far more practical on femtosecond time scales, which require echelon step heights on the order of micrometers rather than millimeters. The advent of two-dimensional (2D) detector arrays and sophisticated computer-based image-processing techniques has enabled the use of two echelons rather than one, thereby greatly increasing the number of time-delay sampling points that can be recorded in a single laser shot (*21*–*23*). Our initial demonstrations of this approach focused on catastrophic sample damage under extreme high-intensity irradiation (*21*, *22*) and, more recently, on reaction dynamics in supercooled liquids and amorphous solids with essentially perfect optical quality and uniformity (*23*). In the present work, through substantial improvements in sensitivity and automated data analysis, we are able to extend single-shot methods to the study of chemical reaction dynamics in homegrown organic crystals. In such samples, the scattered probe light intensity typically exceeded the induced change in probe transmission that was being measured, and the pattern of scattered light at the charge-coupled device (CCD) detector changed appreciably from one crystalline region to another. The entire 20 × 20 grid pattern of transmitted probe beams at the CCD, as well as distortions of the grid pattern, also shifted as a sample crystal was translated because the front and back crystal faces through which the beams passed were not perfectly flat and parallel. To deal with these factors, we developed advanced algorithms in our laboratory for pattern recognition of the 20 × 20 data and reference array grids superimposed over raw images; exclusion of CCD pixels at or near known irregularities (such as grid boundaries); scaled averaging over the roughly 1000 CCD pixels within each grid region that corresponds to a specific delay time; and normalization based on comparison between data and reference grids recorded first without, and then with, the excitation pulse. These measures, taken to improve the sensitivity of the experimental method, reduce the single-shot noise floor to ~0.1% of the transmitted probe light intensity (roughly two orders of magnitude lower than achievable otherwise), thereby enabling single-shot measurements of ultrafast photochemistry in organic crystals.

Transient absorption traces of the aforementioned crystals, each measured in a single laser shot, were acquired at seven probe wavelengths (three of which are shown in Fig. 3) spanning the ground-state I_2^- absorption spectrum (*9*). Each laser shot impinged on a fresh region of the crystal. TPPT, the crystal with the least constrained environment around I_3^-, shows features that are typical of liquid-state triiodide photochemistry (*12*): initial absorption at all probe wavelengths from the I_3^- excited state; formation, after ~400 fs, of I_2^- and I photofragments as evidenced by the characteristic I_2^- absorption spectrum centered near 740 nm; coherent vibrational oscillations of the I_2^- fragment, which by alternately stretching and compressing gives rise to corresponding blue- and red-shifting of its absorption spectrum (fig. S1); and vibrational dephasing and cooling, the former marked by the decay of the coherent oscillations and the latter by a gradual narrowing of the absorption spectrum (observed as substantial decay of absorption at the wings and slower decay or rise of absorption at the center of the spectrum) as the initially excited vibrational population relaxes to the bottom of the I_2^- potential well.

Similar behavior is observed within the first picosecond in the two crystals with more severely constrained environments around the anions: I_3^- excited-state absorption and fragmentation followed by coherent vibrational oscillations of the I_2^- photoproduct. At later times, however, the dynamics differ strikingly from those observed in TPPT or in liquid-state environments. In TBAT, which confines the I_3^- ions within pseudo-columnar channels formed by the tetrabutylammonium counterions, I_2^- absorption decays abruptly and almost completely after 1.1 to 1.3 ps. In the intermediately constrained environment of TEAT, the I_2^- absorption decays 1.4 to 1.7 ps after the initial photolysis, somewhat later and less abruptly than in TBAT. These results suggest substantial lattice environmental effects on the chemistry of the nascent photofragments.

The reaction dynamics were explored further through molecular dynamics simulations. Model systems of 3 × 3 × 3 unit cells were constructed using reported potentials for I_3^- and I_2^- (*14*, *24*, *25*), bond-order potentials (*26*, *27*) for certain covalently bonded counterions, and Lennard-Jones potentials for interactions between other bonded and nonbonded atoms (*9*). A Gaussian thermal distribution of 30,000 trajectories at 298 K was constructed and propagated forward in time in each simulation. Simulations were launched in each model crystal by placing each of the reactive I_3^- ions in a selected electronic excited state (*9*) and then solving the classical equations of motion for each atom; the well-established Verlet algorithm was used to compute new atomic positions and momenta after successive 1-fs time increments. We focus on calculation of I_2^- absorption because this is the experimentally measured property that reveals the lattice-mediated dynamics of key interest. Formation of I_2^- is initiated by defining a "dissociation distance" for the parent I_3^- ion. The first of the two terminal I atoms to move beyond the dissociation distance from the central atom is considered to have dissociated, and the other two I atoms, which remain bonded, are smoothly transferred to the ground-state I_2^- potential-energy surface. The dissociation distance is taken as roughly 115% of the mean equilibrium I_3^- bond length in order to reproduce the temporal onset of I_2^- absorption observed experimentally.

For all three crystals, the simulations reproduce semiquantitatively the onset of I_2^- absorption and coherent vibration. Crucially, the abrupt decay of I_2^- absorption in TBAT and TEAT and the absence of any such decay in TPPT are also reproduced. Individual trajectories selected at random out of the sets of 30,000 show highly consistent outcomes with the respective average trajectories (Fig. 4). The results support the conclusion that the abrupt disappearance of I_2^- spectral signatures arises from lattice reaction cage-mediated recombination of photofragments. In TBAT, the recombination is highly synchronized across the ~10^{12} excited unit cells and confined to a narrow time window of ~200 fs. In TEAT, the recombination is quite well synchronized, occurring mostly within a time window of ~300 fs. In TPPT, however, no signature of synchronized recombination is apparent in either experiment or simulation. In all three crystals, simulations show that the I_2^- fragment moves relatively little after I_3^- photodissociation, whereas the I fragment leaves with about 0.5 eV of kinetic energy, travels a substantial distance (3.3, 4.9, and 3.9 A on average, respectively, in

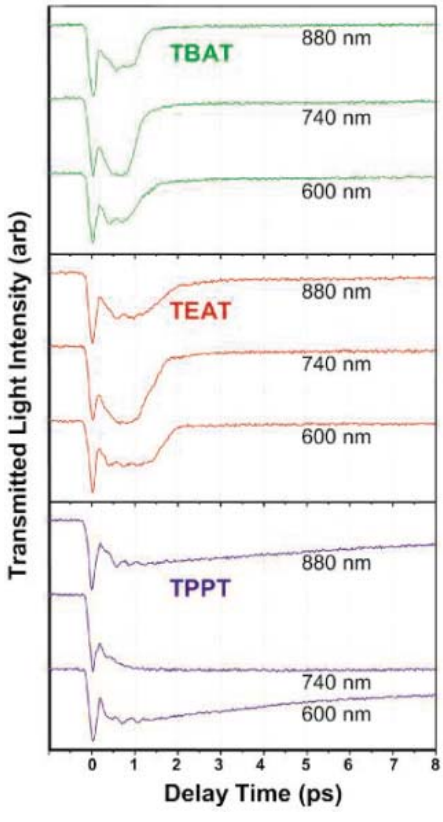
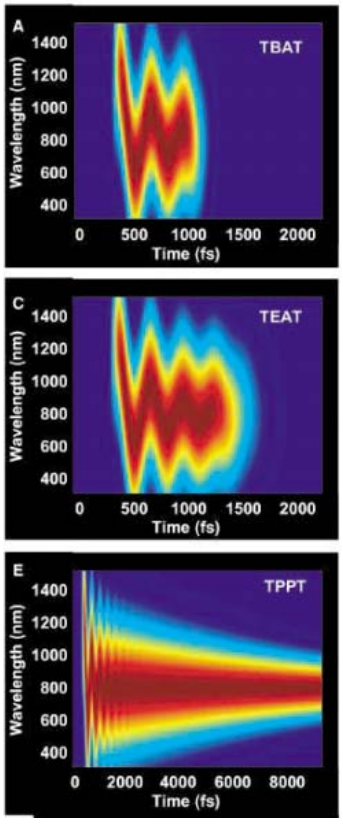


Fig. 3. Single-shot measurements of crystalline chemical reactions. Single-shot data from each of three different crystals are shown at three probe wavelengths. The maximum change in transmitted light intensity is ~2%.

TBAT, TEAT, and TPPT) before its first collision with a neighboring lattice constituent, and loses only a modest amount of its kinetic energy (4, 13, and 13%, respectively) during that encounter. In the TBAT lattice, the dissociated I atom moves away from its parent fragment, remaining within the confines of its pseudo-1D channel, until it collides with a neighboring I_2^- fragment. It then recoils principally along its original path of motion to recombine with its original partner. Although we have conducted classical numerical simulations of the motion, the simulation and experimental results and the particular lattice geometry of TBAT suggest approximate modeling, classically or quantum mechanically, in terms of a particle in a 1D box with rather little dephasing on the time scale of the single round trip that occurs before recombination. Thus, the photofragment's constrained translational motion, from inception through collision to recombination, is substantially phase-coherent. The dynamics are distinct from those observed in photolysis of I_2 in rare gas cryogenic matrices (*8*, *28*), in which energetic molecular states that are dissociative in the gas phase become "cage-bound" by lattice constraints that strongly resist even the initial dissociation event, so there is no free I translation before recoil, which occurs in the molecular electronic excited state but not the ground state. In the present case, a fully dissociated I atom undergoes a rather leisurely excursion whose distance is long compared to the original bond length (2.9 A) and whose duration is long compared to the period during which sudden recombination occurs. In the case of TEAT, this excursion lasts even longer and covers a longer distance. In TEAT, columns of I_3^- ions are oriented in roughly side-on stacks, so that the nearest-neighbor lattice environment for a dissociating I_3^- ion is formed mainly by pseudo-spherical tetraethylammonium ions and adjacent side-oriented I_3^- ions. Our simulations show that different initial conditions among the thermal distribution of I_3^- ions before photoexcitation, particularly different symmetric versus antisymmetric stretching and librational motions, produce different molecular trajectories that give rise to substantial variations in the interactions between a dissociated I atom and its neighbors. This results in recombination that is somewhat less synchronized than in TBAT, in which the local geometry is sufficiently restrictive that similar variations in initial conditions do not appreciably affect the recombination dynamics. Nevertheless, the degree of phase coherence in the recombination of TEAT as well as TBAT, apparent directly from the experimental data as well as from the simulations, is surprising given the complexity of organic crystalline environments generally. The TPPT crystal, whose local lattice surroundings present greatly reduced steric constraints as compared to TBAT and TEAT, permits a wide range of dissociated I-atom trajectories, and both experiment and simulation show no notable recombination during the time window following dissociation.

The experimental and computational results point consistently to a clear correlation between crystalline structure and reaction dynamics. In the cases examined here, well-defined variations in static structure lead to distinct recombination behavior on the part of caged photofragments. Dynamical effects involving transfer of photoproduct translational energy to lattice degrees of freedom, which may permit lattice accommodation of the permanent photoproducts that evidently are formed to some degree in all three crystals, should be studied as a function of temperature. In some systems, dense crystal packing around the reactive species and rapid flow of energy from molecular reactive modes into lattice modes may suppress even the initial photodissociative events. In partially disordered lattices, energy flow from the reactive mode into well-defined degrees of freedom [such as molecular rotation in plastic crystalline phases (*29*)] may suppress reaction through dissipation. This process should be more amenable to incisive analysis than is possible in liquids, which obscure similar phenomena on account of the wide range of ill-defined bath coordinates whose effects generally cannot be examined separately.



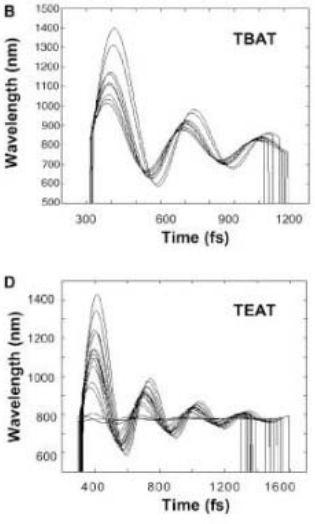


Fig. 4. Molecular dynamics simulations of crystalline chemical reactions. The results are averaged over 30,000 trajectories (left) and for randomly selected individual trajectories (right) for a model of I_3^- photolysis in 3 × 3 × 3 unit cells in (**A** and **B**) TBAT, (**C** and **D**) TEAT, and (**E**) TPPT. The abrupt end of I_2^- absorption in TBAT and TEAT is due to recombination of I_2^- ions with their original I atom partners to regenerate I_3^-. The trajectories shown in (A) and (B) illustrate that recombination is highly synchronized in TBAT and slightly less so in TEAT.

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Anomalous Increase in Carbon Capacitance at Pore Sizes Less Than 1 Nanometer

J. Chmiola,[1] G. Yushin,[1] Y. Gogotsi,[1]* C. Portet,[2]† P. Simon,[2] P. L. Taberna[2]

Carbon supercapacitors, which are energy storage devices that use ion adsorption on the surface of highly porous materials to store charge, have numerous advantages over other power-source technologies, but could realize further gains if their electrodes were properly optimized. Studying the effect of the pore size on capacitance could potentially improve performance by maximizing the electrode surface area accessible to electrolyte ions, but until recently, no studies had addressed the lower size limit of accessible pores. Using carbide-derived carbon, we generated pores with average sizes from 0.6 to 2.25 nanometer and studied double-layer capacitance in an organic electrolyte. The results challenge the long-held axiom that pores smaller than the size of solvated electrolyte ions are incapable of contributing to charge storage.

Supercapacitors, also called electrical double-layer capacitors (EDLCs), occupy a region between batteries and dielectric capacitors on the Ragone plot describing the relation between energy and power (*1*). They have been touted as a solution to the mismatch between the fast growth in power required by devices and the inability of batteries to efficiently discharge at high rates (*2*, *3*). This large capacity for high power discharge is directly related to the absence of charge-transfer resistances that are characteristic of battery Faradaic reactions and subsequently leads to better performance at low temperature. Improvements in the energy density may accelerate the advent of electrical and fuel-cell cars, as well as enable numerous industrial and consumer applications for supercapacitors (*4*). Improvements have been made in cell packaging and electrolytes (*5*, *6*), but a lack of substantial progress in carbon material design has limited energy density, effectively preventing wide-scale usage of supercapacitors.

Unlike batteries and fuel cells that harvest energy stored in chemical bonds, supercapacitors exploit the electrostatic separation between electrolyte ions and high–surface area electrodes, typically carbon (*1*). This results in capacitances of tens of Farads per gram of active material, unlike traditional dielectric capacitors that have capacitances typically measured in microfarads. Energy stored in supercapacitors is linearly proportional to the capacitance of its electrodes, making material optimization crucial.

The large capacitance, C, and hence energy storage potential, of supercapacitors arises due to the small (~1 nm) separation, d, between electrolyte ions and carbon and high (typically 500 to 2000 m^2/g) specific surface area (SSA) of carbon electrodes according to

$$C = \frac{\varepsilon A}{d} \quad (1)$$

where A is the electrode surface area accessible to electrolyte ions, and ε is the electrolyte dielectric constant. Because SSA is explicitly related to pore size, understanding its effect on specific capacitance is especially important and has been the subject of numerous studies over the past decade (*7*–*9*).

Traditional methods of producing porous carbon from either natural precursors such as coconut shell or synthetic precursors such as phenolic resin do not offer sufficient control over porosity (*10*). Mesoporous carbons synthesized by template techniques have produced controllable pores in the range of 2 to 4 nm (*11*). It is believed that pores substantially larger than the size of the electrolyte ion and its solvation shell are required for high capacitance. The use of carbon nanotubes (*12*) has provided a good model system with large pores and high conductivity, leading to impressive power densities but low energy density.

A less well-known class of porous carbons offers great potential for controlling pore size. Carbide-derived carbons (CDCs) are produced by high-temperature chlorination of carbides, whereby metals and metalloids are removed as chlorides, leaving behind nanoporous carbon with a 50 to 80% open pore volume (*13*). Atomic-level porosity control in CDC is achieved by exploiting the host carbide lattice as a template, permitting controlled layer-by-layer metal extraction by optimizing the chlorination parameters. CDCs have a narrow pore-size distribution with a mean value that is tunable with better than 0.05-nm accuracy in the range of ~0.5 to ~3 nm (*14*) and a SSA up to 2000 m^2/g (*15*), which make them attractive candidates for studying porosity in supercapacitor applications. The ease of pore tunability in CDC previously allowed experimental determination of the optimal pore size for hydrogen storage (*16*). Also, CDC has shown impressive specific capacitance when used as the active material in supercapacitors with many electrolyte systems (*17*–*20*). The use of CDC allows precise control over properties found in all carbon materials, allowing broad trends to be discovered that are applicable to other carbons. Previous work with titanium carbide–derived carbon (TiC-CDC) as the active material in supercapacitors with aqueous H_2SO_4 electrolyte (*18*) showed a correlation between the micropore size (pores <2 nm) and capacitance but did not explore pores smaller than 1 nm. This study focused on the small-pore effect by using CDC with pores tuned from 0.6 to 2.25 nm and an electrolyte consisting of a 1.5 M solution of tetraethylammonium tetrafluoroborate in acetonitrile.

TiC-CDC was synthesized by chlorination at 500° to 1000°C (*21*), and its bulk properties were characterized by Raman spectroscopy and transmission electron microscopy (TEM). Conductivity measurements were performed on compacted powders. Porosity was characterized by argon sorption at 77 K and confirmed with data from small-angle x-ray scattering (SAXS) (*22*) and CO_2 sorption at 300 K. The use of multiple techniques for porosity measurement ensures greater confidence in the results. Elec-

[1]Department of Materials Science and Engineering and A. J. Drexel Nanotechnology Institute, Drexel University, Philadelphia, PA 19104, USA. [2]Université Paul Sabatier, CIRIMAT, UMR CNRS 5085, 31062 Toulouse Cedex 4, France.

*To whom correspondence should be addressed. E-mail: gogotsi@drexel.edu

†Present address: Department of Materials Science and Engineering, Drexel University, Philadelphia, PA 19104, USA.

trochemical characterization on two- and three-electrode cells was performed with galvanostatic techniques for measuring capacitance and impedance spectroscopy to measure frequency response. No attempt was made to separate contributions from the positive and negative electrode to detect possible ion sieving because most of the pores were larger than the diameter of the largest unsolvated ion.

TiC-CDC microstructural and porosity development has previously been very well characterized (*18*, *20*), making it an ideal candidate for this study. To determine microstructural development, previous x-ray diffraction (XRD) results have shown that TiC is completely converted to CDC at synthesis temperatures >400°C, and no Bragg peak corresponding to graphite is visible even at a synthesis temperature of 1000°C (*21*). Similar to previous studies, Raman spectroscopy (Fig. 1A) showed a decreasing $R = I_D/I_G$ ratio, the ratio of graphite band (1582 cm^{-1}) intensity to disorder-induced band (~1350 cm^{-1}) intensity, with increasing synthesis temperature, indicating increasing ordering, but as seen previously in XRD, no large-scale graphitization. TEM micrographs (fig. S1, A to C) also showed gradual short-range ordering with rising synthesis temperature. The conductivity increased with synthesis temperature because of a reduction in the concentration of electron scattering defects (Fig. 1A). CDC had a higher conductivity compared to activated carbons from organic precursors with a similar porosity or surface area because it lacked oxygen or hydrogen in its carbon network (*13*).

Several techniques were used to determine the pore size and SSA of CDC. Nonlinear density functional theory (NLDFT) analysis of argon adsorption isotherms (fig. S2A) showed that the width of the pore-size distribution increased with synthesis temperature (fig. S2, B and C), and the average pore size shifted to larger values (Fig. 1B). The pore-size values obtained are in agreement with those obtained via CO_2 sorption at 300 K (fig. S3B) and SAXS analysis (*22*). The BET (Brunauer, Emmet, Teller) SSA showed a similar increase (Fig. 1B). SSA was also calculated by using NLDFT analysis, assuming slit pores, and was later used to corroborate trends revealed with the BET method (fig. S3A). Because the smallest pore size measured by Ar sorption was equal to the unsolvated BF_4^- electrolyte ion size, all the surface area available to the electrolyte ions for charge storage was accessible to Ar. Two advanced activated carbons used commercially in supercapacitors, referred to as NMAC (natural material precursor activated carbon) and SMAC (synthetic material precursor activated carbon), were also studied and served as a reference. They had average pore sizes of 1.45 and 1.2 nm and SSAs of 2015 and 2175 m^2/g, respectively. CDCs synthesized from B_4C and Ti_2AlC (*17*), which have pore sizes of 1.25 and 2.25 nm, respectively, and SSAs of 1850 and 1150 m^2/g, respectively, were also studied because their pore sizes are close to those of typical activated carbons. The results showed that CDC synthesized in the temperature range studied had a pore structure representative of a wide range of activated carbons, making it a good model system to study the effect of pore size on energy storage.

The electrochemical behavior of TiC-CDC is shown in Fig. 2. These results were repeated in experiments at both Drexel University and the University of Paul Sabatier with minimal deviation. The traditional understanding of how porosity affects specific capacitance and frequency response holds that pores larger than the size of the electrolyte ion plus its solvation shell are required for both minimizing the characteristic relaxation time constant, τ_0 (*23*) (the minimum time needed to discharge all the energy from the supercapacitor cell with an efficiency >50%), and maximizing its specific capacitance (*24*). Therefore, because conductivity, surface area, and average pore size all scaled with synthesis temperature, it was expected that CDC synthesized at 1000°C would exhibit the shortest τ_0 and the highest capacitance. Indeed, increasing the average pore size from 0.68 to 1.1 nm caused a slight decrease in τ_0 (Fig. 2A, inset), as expected. Even for the sample with the smallest pore size (500°C TiC-CDC), there was only a minimal decrease in specific capacitance when the current density was increased from 5 to 100 mA/cm^2 (Fig. 2B), which illustrates the minimal change in frequency-response behavior. NMAC and SMAC, which have pore sizes similar to those of TiC-CDC at 1000°C, had time constants similar to those of 800°C TiC-CDC, owing to the higher bulk conductivity of CDC. The opposite trend was found in the behavior of capacitance, however:

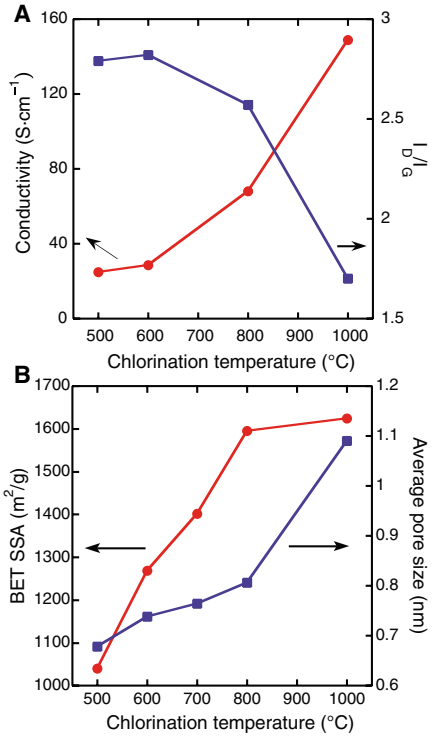

Fig. 1. Effect of synthesis temperature on structure and properties of CDC. The carbon structure (**A**) resolved by Raman spectroscopy showed a decreasing I_D/I_G ratio with increasing synthesis temperature, indicating increasing order. This increasing order was reflected in increasing conductivity with synthesis temperature. Porosity information resolved from gas sorption data shows (**B**) that both the SSA and average pore size increased with synthesis temperature. The calculated error between successive measurements of both pore size and SSA values are within only a few percent.

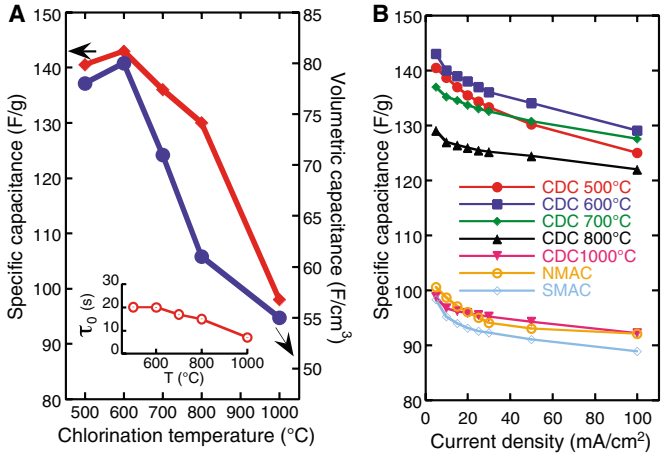

Fig. 2. Electrochemical behavior of TiC-CDC synthesized in the range of 500° to 1000°C. (**A**) Specific capacitance and volumetric capacitance both decreased with synthesis temperature. The maximum error reported in specific and volumetric capacitance was 2.5 and 6%, respectively. Maximum capacitance was at 600°C synthesis temperature. NAMAC and SMAC characteristics are 100 F/g, 35 F/cm^3 and 95 F/g, 45 F/cm^3, respectively, under the same conditions. The plot of characteristic time constant, τ_0, versus synthesis temperature (inset), showed slightly increasing frequency response with temperature. Comparison of TiC-CDC charge-discharge behavior with commercially available carbons (**B**) shows that by using rational design, a 50% improvement can be achieved. There was also very little capacitance fading at current densities up to 100 mA/cm^2, even for the 500°C sample.

Both the specific (gravimetric) and volumetric (capacitance per unit bulk volume of carbon) capacitances decreased with increasing synthesis temperature (Fig. 2A). With an increase in the chlorination temperature from 500° to 1000°C, the specific capacitance decreased by ~40%, from ~140 to ~100 F/g, although the SSA increased by ~60%, from 1000 to 1600 m^2/g. This decrease in capacitance in high–surface area carbons has been attributed to the development of surface area that was inaccessible to electrolyte ions due to the small size of the pores (*25*). In our study, however, the increasing surface area at elevated synthesis temperatures was exclusively the result of larger-diameter pores (Fig. 1B). Therefore, it cannot be explained by the traditional understanding.

When the specific capacitance was normalized by SSA, the effect of pore size, irrespective of surface area, could be ascertained (Fig. 3A). For TiC-CDC, increasing the pore size appeared to have a detrimental effect on the normalized capacitance. Although BET SSA is reported here because it is the most widely used technique and allows direct comparison with data from other studies, specific capacitance was also normalized by DFT SSA, which yielded the same trends (fig. S3A). These results are particularly interesting because the high capacitance of some carbons with pore sizes smaller than 1 nm has been noted before (*9*, *18*, *26*), but a model to explain this behavior has been lacking, and large pores are still considered optimal by most.

Figure 3A shows that there is a trend of decreasing normalized capacitance when the pore size is reduced to ~1 nm, based on data from this study and (*8*, *26*). TiC-CDC synthesized at 1000°C, B_4C-CDC, Ti_2AlC-CDC, NMAC, and SMAC all manifested this behavior, which demonstrated that this size effect was independent of the carbon material used. However, at a pore size below a critical value, as seen with TiC-CDC synthesized below 1000°C, the trend reversed and there was a sharp increase in capacitance with decreasing pore size. Two other carbons with small pores (*8*) follow the same trend.

In region I of Fig. 3A, when pores were substantially larger than twice the size of the solvated ions (Fig. 3B), there was a contribution to capacitance from compact layers of ions residing on both adjacent pore walls. Although the diffuse layer of charge that exists on a planar electrode, classically described by de Levie (*27*), was absent or diminished in size, the capacitance was largely unaffected because the compact layer encompasses much of the potential drop. Decreasing the pore size to less than twice the solvated ion size (Fig. 3C) reduced the normalized capacitance (Fig. 3A, region II) because compact ion layers from adjacent pore walls impinged and the surface area usable for double-layer formation was reduced. This would largely account for the decrease in specific capacitance with pore-size reduction for pore sizes greater than ~1 nm.

This trend reversed with a further decrease in the pore size to less than that of the solvated ion size (Fig. 3D, region III). Decreasing the pore size to a value approaching the crystallographic diameter of the ion led to a 100% increase in normalized capacitance. Dzubiella and Hansen showed that under a potential, there is substantial ion motion and diminished dielectric permittivity in pores less than the size of their solvation shells (*28*). The solvation shell becomes highly distorted as the ion is squeezed through the pore in much the same way a balloon distorts when squeezed through an opening smaller than its equilibrium size. The distortion of solvation shells in small pores of carbon nanostructures was also reported recently (*29*–*31*). Such distortion would allow closer approach of the ion center to the electrode surface, which by Eq. 1, leads to improved capacitance. When the capacitance data from Fig. 3 for pore sizes smaller than the size of the solvated ion (~1 nm) were plotted against the reciprocal of the pore size, a linear relation was obtained (fig. S6). This simplified model, which assumes planar pore surface and constant dielectric permittivity, has important implications. The effects of surface curvature and decreasing dielectric permittivity should decrease the capacitance, which showed the dominance of the 1/*d* term. Whereas templated carbons achieve improved specific capacitance by an increase in the pore size (Fig. 3A, region I, and 3B), resulting in low volumetric capacitance, our model suggests that using microporous carbons with pores smaller than 1 nm allows the volumetric capacitance to increase from 55 to 80 F/cm^3 (Fig. 2A).

The demonstration of charge storage in pores smaller than the size of solvated electrolyte ions will lead to enhanced understanding of ionic transport in porous media. These findings should also permit the design of application-specific supercapacitors: for longer discharge times where energy density is at a premium, such as in hybrid electric vehicles, extremely narrow pores should prove optimal, but for pulse power applications, increasing the pore size might be beneficial. Further tuning the carbon porosity and designing the carbon materials with a large volume of narrow but short pores may allow both energy and power characteristics to be improved.

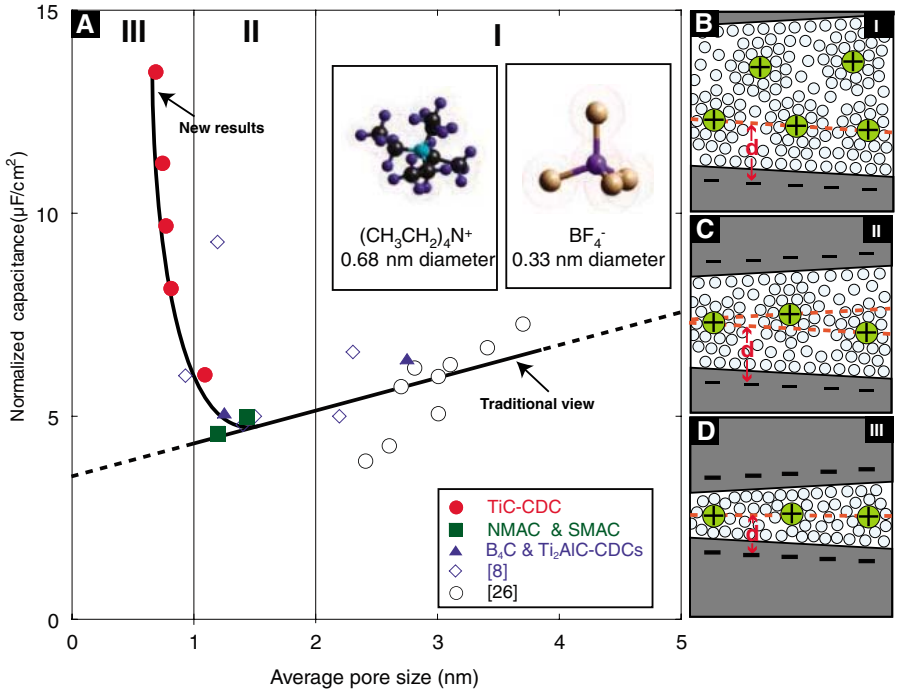

Fig. 3. (**A**) Plot of specific capacitance normalized by BET SSA for the carbons in this study and in two other studies with identical electrolytes. The normalized capacitance decreased with decreasing pore size until a critical value was reached, unlike the traditional view which assumed that capacitance continually decreased. It would be expected that as the pore size becomes large enough to accommodate diffuse charge layers, the capacitance would approach a constant value. (**B** to **D**) Drawings of solvated ions residing in pores with distance between adjacent pore walls (B) greater than 2 nm, (C) between 1 and 2 nm, and (D) less than 1 nm illustrate this behavior schematically.

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Oxygen Isotope Variation in Stony-Iron Meteorites

R. C. Greenwood,[1]* I. A. Franchi,[1] A. Jambon,[2] J. A. Barrat,[3] T. H. Burbine[4]

Asteroidal material, delivered to Earth as meteorites, preserves a record of the earliest stages of planetary formation. High-precision oxygen isotope analyses for the two major groups of stony-iron meteorites (main-group pallasites and mesosiderites) demonstrate that each group is from a distinct asteroidal source. Mesosiderites are isotopically identical to the howardite-eucrite-diogenite clan and, like them, are probably derived from the asteroid 4 Vesta. Main-group pallasites represent intermixed core-mantle material from a single disrupted asteroid and have no known equivalents among the basaltic meteorites. The stony-iron meteorites demonstrate that intense asteroidal deformation accompanied planetary accretion in the early Solar System.

The terrestrial planets formed by the collision and merger of smaller bodies, over a period lasting up to 100 million years (My) after Solar System formation (*1*). The final stage of this process was marked by giant impacts that resulted in large-scale planetary melting (*2*). As a consequence of their protracted formation histories, and subsequent geological reprocessing, the initial stages of planetary accretion are not recorded by these larger bodies. In contrast, asteroids preserve a record of the earliest stages of planetary growth. Thus, W isotopic analysis of iron meteorites (*3*) and Mg isotope studies of basaltic meteorites (*4*) indicate that the earliest asteroids accreted within 1 My of Solar System formation and, due to the presence of live ^{26}Al, rapidly underwent near-total melting.

However, the usefulness of asteroidal material in understanding the earliest stages of planetary formation is hindered by the fragmentary nature of the meteorite record. As a result, we are unable to say how many asteroids are represented in our meteorite collections (*5*). Oxygen isotope analysis is one method that has proven useful in understanding the relationships between the various meteorite sample suites (*6*). The melting event that caused the early-formed asteroids to segregate into a metal-rich core and silicate-rich mantle and crust also homogenized their oxygen isotopes (*7*). The later evolution of these bodies would be by mass-dependent fractionation processes, so that samples derived from the same source asteroid define a single mass fractionation line (slope ≈ 0.52) on a three-isotope diagram (*6*, *8*). Meteorite samples from melted asteroids are collectively referred to as differentiated achondrites.

One limitation with this technique results from the small-scale oxygen isotope variation displayed by many groups of differentiated achondrites (*6*). Thus, main-group pallasites, mesosiderites, and the howardite-eucrite-diogenite suite (HEDs) (*9*) all plot within the same area on an oxygen three-isotope diagram (*6*) and could, in theory, have come from a single asteroid. However, mineralogical and textural evidence indicates that these groups formed in distinct settings, with the HEDs and mesosiderites coming from the outer crust of an asteroid, whereas the main-group pallasites formed deep within their parent body (*10*). Because reflectance spectra indicate that HEDs are from the relatively intact asteroid 4 Vesta (*11*), the main-group pallasites must come from a separate source (*10*). To address the problem of such overlapping oxygen isotope variation, we have undertaken a detailed study of the main-group pallasites and mesosiderites by laser-assisted fluorination (*12*). The high analytical precision of this technique enables the offset between parallel mass fractionation lines to be measured to within ±0.02‰ (*12*) and therefore has the potential to resolve the overlaps seen in the differentiated achondrites.

The results of oxygen isotope analyses (*13*) unambiguously resolve the mesosiderites from the main-group pallasites; thus, each group defines a distinct linear array on an oxygen isotope variation diagram (Fig. 1). The mean Δ^{17}O (*8*) value is –0.183 ± 0.018 (2σ) for the main-group pallasites and –0.245 ± 0.020 (2σ) for the mesosiderites (*13*).

The mean Δ^{17}O value for the mesosiderites is indistinguishable from the previously determined HED value of –0.238 ± 0.014 (*7*). Furthermore, if data from laser-assisted fluorination studies of the HEDs are combined (*7*, *14*), the range in δ^{18}O values for the mesosiderites and HEDs are virtually identical (Fig. 1). It has long been known that the silicate portion of the mesosiderites and the various lithologies of the HED suite show strong mineralogical and geochemical similarities (*10*).

[1]Planetary and Space Sciences Research Institute, Open University, Walton Hall, Milton Keynes, MK7 6AA UK. [2]Laboratoire Magmatologie et Géochimie Inorganique et Expérimentale, Université Pierre et Marie Curie, CNRS UMR 7047 case 110, 4 place Jussieu, 75252 Paris Cedex 05, France. [3]Université de Bretagne Occidentale–Universitaire Européen de la Mer, CNRS UMR 6538 (Domaines Océaniques), place Nicolas Copernic, F-29280 Plouzané Cedex, France. [4]Department of Astronomy, Mount Holyoke College, South Hadley, MA 01075, USA.

*To whom correspondence should be addressed. E-mail: r.c.greenwood@open.ac.uk

This evidence, combined with their similar oxygen isotope variation, suggests that both groups may have a common origin. However, a single source for these groups is not widely accepted, objections cited being (i) lack of remote sensing evidence that mesosiderites are present on Vesta (*15*), (ii) lack of metal-rich clasts in howardites (*10*, *16*), (iii) paucity of olivine in the HEDs (*10*), and (iv) silicate clast populations in the two groups that are not identical, although geochemically similar (*10*, *16*).

On closer scrutiny, these arguments are less persuasive. Remote sensing does not indicate the presence of mesosiderites on Vesta, simply because metallic iron does not have any spectral features in the visible and near-infrared. Howardites containing mesosiderite-like clasts have now been identified (*17*) and, in addition, platinum-group element signatures in both brecciated eucrites and mesosiderites show strong similarities (*18*). Paucity of olivine in HEDs is not a strong argument in favor of separate parent bodies. Olivine is an accessory phase in HEDs (*10*), and olivine-rich diogenites, with up to 45% modal olivine, have now been identified (*19*). It is well understood that the metal-silicate mixing event that formed the mesosiderites took place at high temperatures, with the metal fraction being essentially molten (*20*). Comparing these altered mesosiderite clasts with unaltered HED material is not in itself a compelling argument for separate source asteroids (*10*, *16*), because local-scale metal-silicate mixing processes on a single parent body could equally explain these differences.

Vesta has been proposed as the parent body of the HEDs, mainly due to its distinctive reflectance spectrum, which is close in structure to the laboratory-measured HED spectra (*10*, *11*). The link between Vesta and the HEDs has been strengthened by the discovery of smaller asteroids known as Vestoids, with Vesta-like spectra and apparently derived from it by impact processes (*11*). A number of Vestoids occupy positions in the asteroid belt between Vesta and the 3:1 meteorite-supplying resonance (~2.5 astronomical units) and hence solve the problem of how fragments of Vesta can be placed into Earth-crossing orbit (*11*). If mesosiderites are also derived from Vesta, they are likely to be exposed at the asteroid's surface and should be detectable using the instrument package on the NASA Dawn discovery mission (*21*).

Recent isotopic dating is broadly consistent with a single source for the HEDs and mesosiderites. Mg isotopes indicate that initial formation of the HEDs and mesosiderites was contemporaneous, occurring <1 My after Solar System formation (*4*). Evidence that Mn/Cr fractionation in the Vaca Muerta mesosiderite took place ~2 Myr later than in the HEDs (*20*) appears to be at odds with evidence that Al/Mg fractionation occurred slightly later in the HEDs than in Vaca Muerta (*4*). These differences may reflect isotopic disturbance during metal-silicate mixing (*4*, *20*). There is also uncertainty concerning the timing of the metal-silicate mixing event, with Cr isotopes indicating a younger age (>20 My after Solar System formation) than W isotopes (~5 My after Solar System formation) (*20*). This again may result from isotopic disturbance during metal-silicate mixing (*20*).

The origin of mesosiderites remains controversial, with a diverse range of models put forward to explain their genesis (*15*, *16*, *20*, *22*, *23*). They may have formed when the molten metal core of a denuded asteroid impact-mixed with the outer layers of a second differentiated body (*16*, *20*, *22*). Alternatively, the mesosiderites are viewed as the products formed when an asteroid with a molten core was disrupted and subsequently reaccreted (*23*). If mesosiderites and HEDs are both derived from the relatively intact asteroid Vesta, total parent-body disruption and subsequent reaccretion appears unlikely (*23*). Thus, impact of a molten asteroid core into the surface layers of a second differentiated asteroid is the more plausible model to explain mesosiderite formation (*16*, *20*, *22*). Evidence indicating that mesosiderites cooled rapidly at high temperatures (~10^5°C/My in the range 850° to 1150°C) (*23*), but slowly at lower temperatures (~0.03° to 1°C/My in the range 400° to 250°C) (*23*), possibly due to the formation of an ejecta blanket, appears compatible with this model.

Close encounters between asteroids would have been common events in the early Solar System and could result in the smaller of the bodies being left in a deformed state, having lost much of its crust and mantle (*24*). Pressure release associated with such gravitational unloading may have caused remelting of the remaining core (*24*). Given the extent of collisional encounters in the early Solar System (*1*, *24*), processes similar to those that formed the mesosiderites must have been relatively commonplace events. The unique aspect of the mesosiderites is that the evidence for these processes has been preserved and not obliterated by later collisions and mergers; this is possibly because their formation was a relatively late-stage event (*20*).

It is clear from the results of this study that the main-group pallasites are derived from an asteroidal source distinct from that of either the mesosiderites or the HEDs (Fig. 1). The main-group pallasite mean $\Delta^{17}O$ value of –0.183 ± 0.018 (2σ) is unique and does not correspond to any known group of basaltic meteorites. Main-group pallasites must therefore sample a disrupted differentiated asteroid, in which much of the outer crustal material was removed.

There is general agreement that main-group pallasites are samples from the core-mantle boundary of a differentiated asteroid (*25*, *26*). Because metal sinks to form the core of a molten asteroid and olivine would be the major constituent of the lower mantle, this model appears reasonable (*10*). However, two outstanding problems remain: (i) main-group pallasites seem overabundant relative to the small volume of the core-mantle interface and (ii) because of their large difference in density, olivine and liquid metal should unmix rapidly (*10*).

The primary angular olivine texture of main-group pallasites (*25*, *26*) is generally regarded as the product of a major impact event, in which olivine-rich lower mantle material was catastrophically driven into the underlying partially molten core (*10*, *25*, *26*). This was followed by rapid solidification of the metal melt, thus preventing any subsequent unmixing (*25*, *26*). In consequence, pallasites should no longer be regarded as mere samples of an asteroid's core-mantle boundary but rather as an impact-generated lithology composed of intermixed core and mantle material. This explains the apparent overabundance of main-group pallasites (*10*), because an impact-generated mixing zone would have a much greater volume than a gravity-produced core-mantle boundary layer,

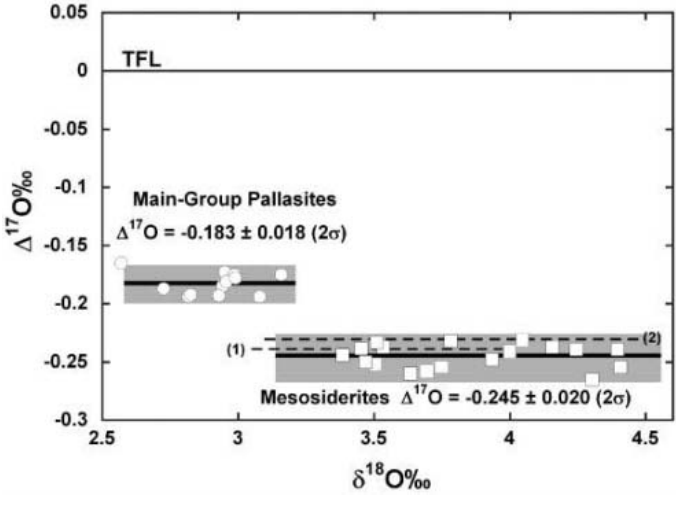

Fig. 1. Oxygen isotope variation in mesosiderites and main-group pallasites. Analytical data for individual samples is given in table S1. Solid lines show the mean $\Delta^{17}O$ value for both the mesosiderites and main-group pallasites. For clarity, error bars for individual analysis have been omitted. However, the gray-shaded boxes depict the 2σ error on the mesosiderite and main-group pallasite population $\Delta^{17}O$ and $\delta^{18}O$ mean values. Open squares, mesosiderite analyses; open circles, pallasite analyses; TFL, terrestrial fractionation line. Line 1 is the eucrite fractionation line of (*7*); line 2 shows the range of HED values obtained by (*14*).

which is essentially just a two-dimensional interface. The lack of basaltic meteorites with a $\Delta^{17}O$ value equivalent to the main-group pallasites suggests that the outer crustal layers were removed from the parent asteroid. This may have taken place during the pallasite-forming impact event itself. If loss of surface material took place as the result of a collisional encounter (*24*), the extent of silicate loss appears to have been much less in the pallasites than mesosiderites, because the former show no evidence for remelting of the metal core.

Metal in main-group pallasites is compositionally similar to that in the IIIAB irons, such that both groups are probably derived from the same asteroid (*26*). W isotope dating of IIIAB irons indicates that the metal-silicate segregation event that formed them occurred extremely early (<1 My after Solar System formation) (*3*). Mn-Cr dating is consistent with an early formation age for main-group pallasites (*10*), although the isotope systematics show evidence of disturbance, probably in part as a consequence of impact-related processes (*27*). The cooling rates recorded by main-group pallasites also reflect impact-related processes. Thus, cooling at high temperatures (~1100°C) took place 10^6 times faster than at lower temperatures (~1°C/My at 800° to 400°C) (*27*). The higher rates may have resulted from impact-driven mixing of partially molten metal and solid silicate mantle material, whereas slower cooling may reflect postimpact burial (*25*).

The results presented here demonstrate that pallasites and mesosiderites are derived from distinct asteroidal sources. Like the HEDs, mesosiderites are probably samples from the asteroid 4 Vesta, a conclusion that can be tested by the NASA Dawn Mission (*21*). It is also clear from these results that extensive asteroidal deformation was an important process during the early stages of planetary formation. Recent observations suggest that such collisional reprocessing of planetesimals may also be a significant feature of extrasolar planetary systems, such as the solar-type star BD+20 307 (*28*).

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9. Main-group pallasites and mesosiderites are composed of subequal amounts of Fe-Ni metal and silicate material. The two groups are texturally and compositionally quite distinct. Main-group pallasites consist of large olivine crystals enclosed in Fe-Ni metal, whereas mesosiderites are complex breccias of metal-rich clasts, basaltic, gabbroic, and rarer pyroxenitic fragments enclosed in a fine-grained metal-silicate matrix. HEDs are a suite of basaltic and related coarser-grained igneous meteorites that are believed to be samples of the outer crust of asteroid 4 Vesta (*10*).
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Supporting Online Material

www.sciencemag.org/cgi/content/full/1128865/DC1
Materials and Methods
Figs. S1 and S2
Table S1

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Self-Healing Pulse-Like Shear Ruptures in the Laboratory

George Lykotrafitis, Ares J. Rosakis,* Guruswami Ravichandran

Models predict that dynamic shear ruptures during earthquake faulting occur as either sliding cracks, where a large section of the interface slides behind a fast-moving rupture front, or self-healing slip pulses, where the fault relocks shortly behind the rupture front. We report experimental visualizations of crack-like, pulse-like, and mixed rupture modes propagating along frictionally held, "incoherent" interfaces separating identical solids, and we describe the conditions under which those modes develop. A combination of simultaneously performed measurements via dynamic photoelasticity and laser interferometry reveals the rupture mode type, the exact point of rupture initiation, the sliding velocity history, and the rupture propagation speed.

A central issue in the modeling of earthquake rupture is the duration of slip at a point on the fault as compared to the duration of the rupture of the entire fault (*1*). In the classical crack-like mode of shear rupture (*2*), the slip duration at a point (rise time) is a considerable fraction of the overall rupture propagation time. In other words, the duration of the ground shaking, witnessed locally by an observer, is comparable to the overall duration of the earthquake. This mode has been generated in several numerical simulations of spontaneous rupture, when a rate-independent friction law was implemented (*3*–*7*). However, inversions of seismic data for slip histories from well-recorded events indicate that the duration of slip at a point on the fault is often one order of magnitude shorter

Graduate Aeronautical Laboratories, California Institute of Technology, Pasadena, CA 91125, USA.

*To whom correspondence should be addressed. E-mail: Rosakis@aero.caltech.edu

than the event duration (*8*–*10*). This observation has led to the concept of pulse-like rupture modes (*8*, *11*), in which the slip is confined to a finite distance behind the propagating rupture front while the fault continuously locks.

Various mechanisms for self-healing pulse generation along a homogeneous fault have been proposed. One such mechanism suggests that if the fault strength is low immediately behind the rupture front and if it increases rapidly at a finite distance, resulting in a robust join between the contacting surfaces, then the slip would be restricted to a short and narrow propagating area (*12*). For example, a strong velocity-weakening friction law model (that is, the fault strength drops rapidly with sliding speed) could indeed allow, under certain conditions, for a pulse-like behavior of rupture to develop. Such models are supported by recent laboratory experiments with high sliding rates (*13*). However, simulations using velocity weakening have sometimes resulted in crack-like propagation or self-healing pulse-like propagation (*14*–*20*). Friction laws operating along faults that separate two identical elastic solids have to include laboratory-based rate and state evolution features (such as dependence on the slip rate and on the history of fault evolution), although these laws should not induce ill-posedness or paradoxical features (*20*) of the mathematical model of non-uniform sliding. It has been proven that generalized rate and state friction laws are appropriate candidates for modeling rupture in uniform faults (*20*–*22*). For rupture to occur as a self-healing pulse (*1*, *22*), three requirements have to be fulfilled. First, the friction law must include strengthening with time on slipped portions of the fault that are momentarily in stationary contact (*16*). Second, the velocity weakening at high slip rates must be much greater than that associated with the weak logarithmic dependence observed in the laboratory during low-velocity sliding experiments. Third, the overall driving stress has to be lower than a certain value but high enough to allow for self-sustained pulse propagation (*22*).

Strong velocity weakening may explain the onset of short-duration slip pulses along faults that separate similar materials. Other mechanisms exist as well. One mechanism (the barrier model) involves the geometric confinement of the rupture domain by unbreakable regions. In that case, an earthquake consists of a number of short-duration crack-like ruptures on a small rupture area that are separated by locked regions (*5*, *23*). Alternatively, the rupture may nucleate and propagate in both directions along the fault, while one of its tips arrests suddenly at a strong barrier in one side of the hypocenter. Consequently, after its arrest, the reflected waves from the barrier spread back and heal the rupture surface. In this case, the pulse-like configuration results from the interaction of the still-propagating end of the rupture with the healing reflected wave (*24*).

The variations in normal stress on the rupture interface are also thought to be responsible for pulse formation (*25*, *26*). As an example, the expansion of pore fluid, caused by frictional heating, may dramatically reduce the effective normal stress and consequently the frictional resistance. Pore pressure, however, may also rapidly decrease behind the rupture tip, causing a restrengthening and possibly a locking of the fault.

We performed a number of well-controlled experiments, all of which involved dynamic sliding along "incoherent" (frictional) interfaces separating identical materials (that is, homogeneous systems), to investigate the generation of the two rupture modes described above and to confirm the existence of pulse-like ruptures. Two plates of Homalite-100, subjected to a far-field uniform compressive stress, were frictionally held along the interface (or fault). The top plate was also subjected to dynamic shear loading. All the experiments were executed at the same external confining stress of P = 10 MPa, applied by a calibrated press. Dynamic photoelasticity, depicting the full-field contours of maximum shear stress, was combined with a laser interferometry–based technique, giving an accurate local measurement of the sliding velocity at the interface within an experimental error on the order of 1% (*27*). Detailed descriptions of the experimental configuration and the techniques used are provided in the supporting online material (SOM) (fig. S1).

In an instantaneous isochromatic fringe pattern obtained at an impact velocity V of 19 m/s (Fig. 1A), an eye-shaped fringe structure was observed traveling behind the longitudinal wavefront from right to left. The rupture tip A followed this fringe structure at a supershear speed of 1.36 C_S, where C_S is the shear wave speed of Homalite. Consequently, two Mach lines of shear stress discontinuity (*28*), forming a shear Mach cone, emanated from the sliding tip. The rupture tip speed v, which was obtained by two methods, was found to be constant. In the first method, the position of the tip was followed in various frames and found to be well approximated as a linear relation, giving v. In the second method, the Mach angle θ was measured and, by means of the relation $v = C_S/\sin\theta$ (*28*), v was obtained frame by frame.

The high-speed camera and both interferometers were simultaneously triggered. The synchronization of the 16 images captured by the high-speed camera, with the horizontal component of the local relative particle velocity recorded by the velocimeters, allowed the correlation of the characteristic features appearing in the photoelastic image (Fig. 1A) with the relative velocity history diagram (Fig. 1B). When the longitudinal wavefront arrived at the velocity measurement positions M_1 and M_2 (where the pair of the interferometric velocimeters was pointed), the velocities of both points started to increase. However, the relative horizontal velocity (Fig. 1B) was zero for the next few microseconds, and it remained very low for a time interval of approximately 13 μs. A numerical integration of the relative velocity with respect to time from 0 to 13 μs resulted in a net relative horizontal displacement of 2 μm between points M_1 and M_2 (fig. S4B). This observation is explained as an elastic shear deformation between M_1 and M_2, rather than rupture (interfacial sliding). The dashed line in Fig. 1B establishes the relative horizontal velocity level that corresponds to sliding initiation for this particular experiment. At approximately 13 μs, the relative velocity (Fig. 1B) rose sharply, and interfacial sliding began. After it reached its maximum of ~6 m/s, the relative velocity decreased and then fluctuated but never fell below 4 m/s during the recording time. The sliding was continuous, and thus we can safely say that rupture occurred in a classic crack-like mode. Additional details are provided in the SOM (figs. S3 and S4).

v was substantially higher than C_S, which resembles supershear rupture propagation occurring along "coherent" interfaces separating identical monolithic solids (*28*–*30*). However, in contrast to previous studies (*28*, *30*), the present work involves incoherent (or frictional) interfaces and a static far-field compressive loading. Here, the frictional resistance to sliding depends

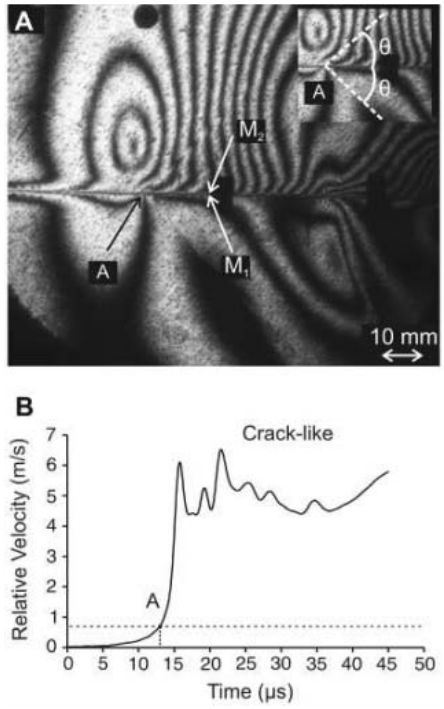

Fig. 1. (**A**) Isochromatic fringe pattern generated during an experiment in which the impact speed was 19 m/s. The rupture tip is at the fringe concentration point A. The inset highlights the location of the Mach lines emanating from the rupture tip and the specimen configuration. (**B**) Relative velocity history of points M_1 and M_2, which belong to the upper and lower plate, respectively; the two points are located at a distance of 70 mm from the impact side of the Homalite plates. The rupture commenced when the rupture tip A reached the velocity measurement position.

on the normal stress through the friction law. The fault-normal stress, however, is a superposition of the static externally imposed pressure and a dynamic (inertial) compression generated as follows: The pressure wave produced by the impact loading creates a primarily horizontal compressive stress in the upper plate close to the frictional interface. As a result of the Poisson effect (in which compression in the horizontal direction causes expansion in the vertical direction), the pressure wave also creates compression in the direction that is vertical to the interface. As the sliding proceeds, the vertical stress to the rupture interface changes, and thus the frictional resistance also changes. Consequently, we infer that sliding depends on impact loading, not only through the asymmetric horizontal compression, which is the driving force for sliding, but also through the vertical compression, which affects the resistance to sliding. Such dependence causes essential changes in the rupture modes that are expected to occur as the impact speed decreases.

Indeed, the reduction of the impact speed from 19 to 17 m/s resulted in drastic changes in the fringe pattern geometry (Fig. 2A). It also changed the horizontal relative velocity history of points M_1 and M_2 (Fig. 2B). By synchronizing the captured photoelastic frames with the horizontal relative velocity history, we were able to correlate the fringe concentration points A_1 and A_2 with abrupt changes in the horizontal relative velocity (Fig. 2B). At A_1, the relative velocity increased rapidly and the sliding started. This observation signifies that A_1 was the rupture tip. Thereafter, the relative velocity oscillated up to point A_3 (Fig. 2A), where the relative velocity increased and remained high for the rest of the recording time. The propagation speeds of A_1 and A_2 were 1.19 C_S and 1.0 C_S, respectively. Point A_3 propagated at a sub-Rayleigh speed of 0.85 C_S and signified the start of a clearly crack-like behavior of the rupture. Thus, it can be concluded that the event of rupture was constituted by two distinctive modes. From A_1 to A_3, the relative velocity changed twice, forming two pulses that were followed by a crack-like rupture mode that commenced at A_3.

By further decreasing the impact speed, a similar, albeit much simpler, behavior of the relative velocity was observed. Once again, we identified two rupture tips, A_1 and A_2, which were fringe concentration points and propagated along the rupture interface at speeds of 1.09 and 0.98 C_S, respectively (Fig. 2, C and D). The initial relative deformation at the velocity measurement position was elastic shear until approximately 18 μs, when the rupture tip A_1 arrived there and sliding commenced. As in the previous cases, the commencement of slip corresponded to an accumulated relative horizontal displacement of 2 μm. Subsequently, the horizontal relative velocity increased rapidly from 0.7 m/s to a local maximum of 2.5 m/s at 20 μs. After 5 μs, the relative velocity decreased abruptly back to 0.7 m/s at point A_2 (Fig. 2D). Because the relative velocity was very low, the slip ceased, allowing surface asperities to reestablish contact and be deformed elastically. Below the level established by the dashed line (Fig. 2D), we assume that there is no sliding and that the recorded velocity is only due to elastic shear deformation between points M_1 and M_2. This conjecture is strengthened by the relative displacement history (fig. S5B), which shows that, from 24 until 25 μs, the relative displacement was almost constant. These observations show that the stable fringe structure (A_1A_2) represents a self-healing slip pulse of ~7 μs in duration. Directly after the pulse, the relative velocity increased rapidly to 6.4 m/s and retained its large value of ~4 m/s for a relatively long period of time (~40 μs). The sliding velocity structure suggests that the initial rupture of the pulse-like mode was immediately followed by a second rupture of the crack-like mode. Thus, the experimental results indicate that the rupture process is very sensitive to impact speed. Indeed, as the impact speed was decreased while the external confining stress was kept constant, the rupture mode changed from a crack-like mode to a mixed mode, where either multiple slip pulses or a single self-healing slip pulse was followed by a crack.

The formation of pulses (Fig. 2) cannot be explained by the barrier model. A very simple calculation (fig. S2) reveals that these pulses were formed well before the arrival of the reflected waves (from the top and bottom hori-

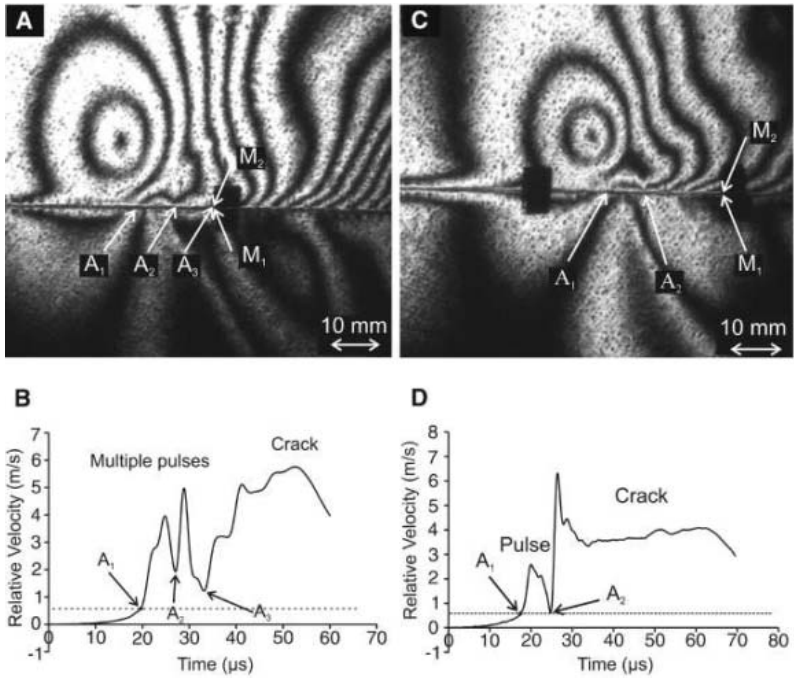

Fig. 2. (**A**) Isochromatic fringe pattern generated during an experiment in which the impact speed was 17 m/s. (**B**) Relative velocity history of points M_1 and M_2 located at a distance of 70 mm from the impact side of the Homalite plates. Two pulses, A_1A_2 and A_2A_3, were formed. The crack-like rupture mode initiated at A_3 immediately behind the second pulse. (**C**) Isochromatic fringe pattern generated during an experiment in which the impact speed was 13 m/s. (**D**) Relative velocity history of points M_1 and M_2 located at a distance of 30 mm from the impact side of the Homalite plates. A self-healing pulse A_1A_2 was formed. The crack-like rupture mode initiated at A_2 immediately behind the second pulse.

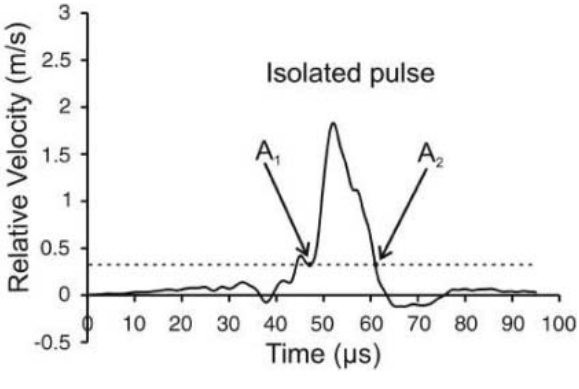

Fig. 3. Relative velocity history of points M_1 and M_2 for an experiment in which the impact speed was 10 m/s. M_1 and M_2 were located at a distance of 70 mm from the impact side of the Homalite plates. An isolated pulse A_1A_2 was formed.

zontal surfaces of the Homalite plates that were blocked by the hydraulic press) to the velocity measurement position. Also, the free surfaces (across the smallest dimension of the Homalite plates) did not act as barriers. We conjecture that the pulse formation was due either to the velocity-weakening character of the friction law or to the changes in the frictional resistance caused by non-uniform variations in dynamic normal stress on the rupture interface, or to a combination of both phenomena.

When the impact speed was further reduced to 10 m/s, the rupture mode became purely pulse-like (Fig. 3). The rupture started at A_1 and propagated at a sub-Rayleigh speed of 0.76 C_S, whereas, after 15 μs, the sliding ceased at A_2. The duration of sliding was very short compared to the ~100-μs duration of the impact event. Thus, we infer that an isolated pulse was formed. Such a case clearly indicates that a purely pulse-like mode of rupture can occur under the appropriate conditions.

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Materials and Methods
Figs. S1 to S5
Reference

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Observations of Biologically Generated Turbulence in a Coastal Inlet

Eric Kunze,[1]* John F. Dower,[1]* Ian Beveridge,[2] Richard Dewey,[1] Kevin P. Bartlett[1]

Measurements in a coastal inlet revealed turbulence that was three to four orders of magnitude larger during the dusk ascent of a dense acoustic-scattering layer of krill than during the day, elevating daily-averaged mixing in the inlet by a factor of 100. Because vertically migrating layers of swimming organisms are found in much of the ocean, biologically generated turbulence may affect (i) the transport of inorganic nutrients to the often nutrient-depleted surface layer from underlying nutrient-rich stratified waters to affect biological productivity and (ii) the exchange of atmospheric gases such as CO_2 with the stratified ocean interior, which has no direct communication with the atmosphere.

Turbulent mixing in the ocean plays key roles in a wide range of processes, from regulating the large-scale thermohaline overturning circulation (also known as the global conveyor belt) and water-mass modification, to the dispersal and dilution of anthropogenic waste. Below the surface mixed layer, turbulent mixing controls the exchange of water properties between the surface layer, which is in direct contact with the atmosphere, and the density-stratified ocean interior, where mixing is typically reduced to diffusivities on the order of 0.1×10^{-4} m^2 s^{-1} (*1*, *2*), controlled by the breaking of internal waves generated by the wind and tides. Exchange across the highly stratified base of the surface mixed layer influences not only biological productivity through nutrient supply but also air/sea gas exchange (*3*). By providing another mechanism by which nutrients and tracers can pass between the nutrient-limited surface mixed layer and the underlying nutrient-replete stratified ocean, biologically generated turbulence could (i) explain how surface production is often higher than can be accounted for by known mixing mechanisms (*4*) and (ii) regulate gas exchange between the ocean and atmosphere, which plays a key role in the carbon cycle, carbon sequestration, and climate.

Although marine organisms have long been known to be capable of generating turbulence (*5*–*8*), the role of biologically generated turbulent mixing in the ocean has largely been neglected, perhaps because this mechanism was discounted in earlier work (*9*). However, more recent evaluations based on the energetics of swimming organisms suggest that species ranging in size from large zooplankton (0.5 cm) to cetaceans on the order of 10 m long can generate turbulent dissipation rates ε (the rate at which turbulent kinetic energy is damped by molecular viscosity) on the order of 10^{-5} W kg^{-1} within schools and swarms (*10*, *11*). Such high values, fully three to four orders of magnitude larger than average turbulence levels in the stratified ocean, have the potential to dominate mixing in the upper ocean, where marine organisms are most abundant. Here we report observations quantifying biologically generated turbulence in a coastal inlet.

The above notion was tested by collecting microstructure profiles with depth during dusk in Saanich Inlet, British Columbia. The profiler measures microscale (1 cm) shear, temperature, and conductivity, as well as fine-scale (1 m) temperature, conductivity, and pressure. Profiles were collected at 3-min intervals. Microscale shear measurements were de-spiked before processing to remove plankton collisions. Following common practice, dissipation rates were estimated by iteratively fitting shear spectra from 4-m half-overlapping profile segments to a turbulence model spectrum (*12*). The wavenumber band from 1 to 100 cpm (wavelengths of 0.01 to 1 m) was typically fit for dissipation rates exceeding 10^{-7} W kg^{-1}. A shipboard 200-kHz ASL Environmental Sciences Water Column Profiler single-beam echosounder (*13*)

[1]School of Earth and Ocean Sciences, University of Victoria, Post Office Box 3055, STN CSC Victoria, BC, V8W-3P6, Canada. [2]Department of Biology, University of Victoria, Post Office Box 3020, STN CSC Victoria, BC, V8W-3N5, Canada.

*To whom correspondence should be addressed. E-mail: kunze@uvic.ca (E.K.); dower@uvic.ca (J.F.D.)

tracked the vertical movement of a dense acoustic-scattering layer of euphausiids. The echosounder transmitted 300-μs pulses at 1 Hz. Volume backscatter intensity was recorded in 0.125-m depth bins.

Saanich Inlet is a semi-enclosed fjord with a maximum depth of about 240 m. It is connected to the Strait of Georgia through narrow tidal channels with a 70-m sill. Typical turbulent dissipation rates in the inlet are less than 10^{-9} W kg^{-1} (*14*). Waters below about 100 m depth are anoxic because of bacterial decomposition of settled biomass. Despite weak winds and tides, the inlet is very productive in the summer. The supply of nutrients for this productivity is thought to arise from tidal mixing outside the inlet, followed by advection into the inlet by tidally rectified mean currents. This hypothesis is supported by a 14-day cycle in nutrients and blooms (*14*).

Saanich Inlet hosts a large resident population of the euphausiid (krill) *Euphausia pacifica*, which occur in concentrations of up to 10,000 individuals m^{-3}; that is, one krill every 5 cm (*15*, *16*). *E. pacifica* is the most common euphausiid in the coastal waters of the northeast Pacific and is an important prey item for many fish species, including herring (*17*). It is also a strong vertical migrator. During daylight, euphausiids in Saanich Inlet remain largely sedentary, resting just above the anoxic interface at about 100 m, presumably to hide from visual predators (*18*). As dusk approaches, the euphausiids swim toward the surface, with the smallest individuals preceding the largest 2-cm-long individuals by approximately 20 min (*19*). Laboratory studies have shown that, at peak swimming speeds, individual *E. pacifica* generate a jet with maximum speeds of about 5 cm s^{-1} (*20*). Acoustic observations of the *E. pacifica* population in Saanich Inlet suggest average ascent speeds of 2.5 to 3.5 cm s^{-1} (*21*). At dawn, the largest individuals begin the descent, followed by smaller individuals. This size bias in the timing of ascent and descent has been interpreted as further evidence of predator avoidance; smaller animals ascend earlier and descend later because they are less visible to predators than are larger animals (*22*, *23*).

Our measurements took place during late April 2005. Winds were very light throughout. Waters below 5 m depth were strongly stratified, with buoyancy frequencies of 10^{-2} rad s^{-1}. Even higher stratifications were found above, with well-mixed surface layer thicknesses not exceeding 3 m. During daylight, the dense backscattering layer of krill remained stationary at about 100 m, and turbulent dissipation rates ε were close to the instrument noise level of 10^{-9} W kg^{-1} (Fig. 1), which is comparable to signals typically found in the open ocean (*1*). At dusk on 28 April, the backscattering layer began to ascend and become more diffuse. Initially, no enhanced microstructure was observed. However, as the base of the scattering layer [thought to be associated with larger euphausiids, with Reynolds numbers sufficiently high to generate turbulence (*10*)] began to shoal, turbulence levels between 30 and 100 m depth approached 10^{-5} to 10^{-4} W kg^{-1} for a 10- to 15-min interval. These values are 100 to 1000 times the dissipation rates associated with turbulence patches in the stratified deep ocean and are comparable to values found in strongly turbulent tidal channels (*1*, *24*). Corresponding diffusivities during this interval were 200×10^{-4} m^2 s^{-1} to 2000×10^{-4} m^2 s^{-1}. Density remained well stratified during the measurement interval, so turbulence cannot be attributed to mixed-layer deepening; during the sampling interval, 3-m temperature decreased by 3°C because of surface cooling or horizontal advection, but this surface-intensified cooling was confined to a depth of 15 m or less. The microstructure shear spectra produced resemble those associated with shear-driven turbulence, and unstable density overturns of 1 to 10 m during this interval are consistent with the above dissipation rates. This is at odds with the 1.5-cm length of individual krill, suggesting that they act in concert rather than as individuals when they swim upward. By acting communally, they would reduce viscous drag.

The backscattering layer ascended 100 m in less than 15 min, which is consistent with the swimming speeds recorded in the lab of 5 to 10 cm s^{-1} (*20*). Because the period of peak ascent lasted less than 15 min, it is not difficult to imagine how it could have been undersampled

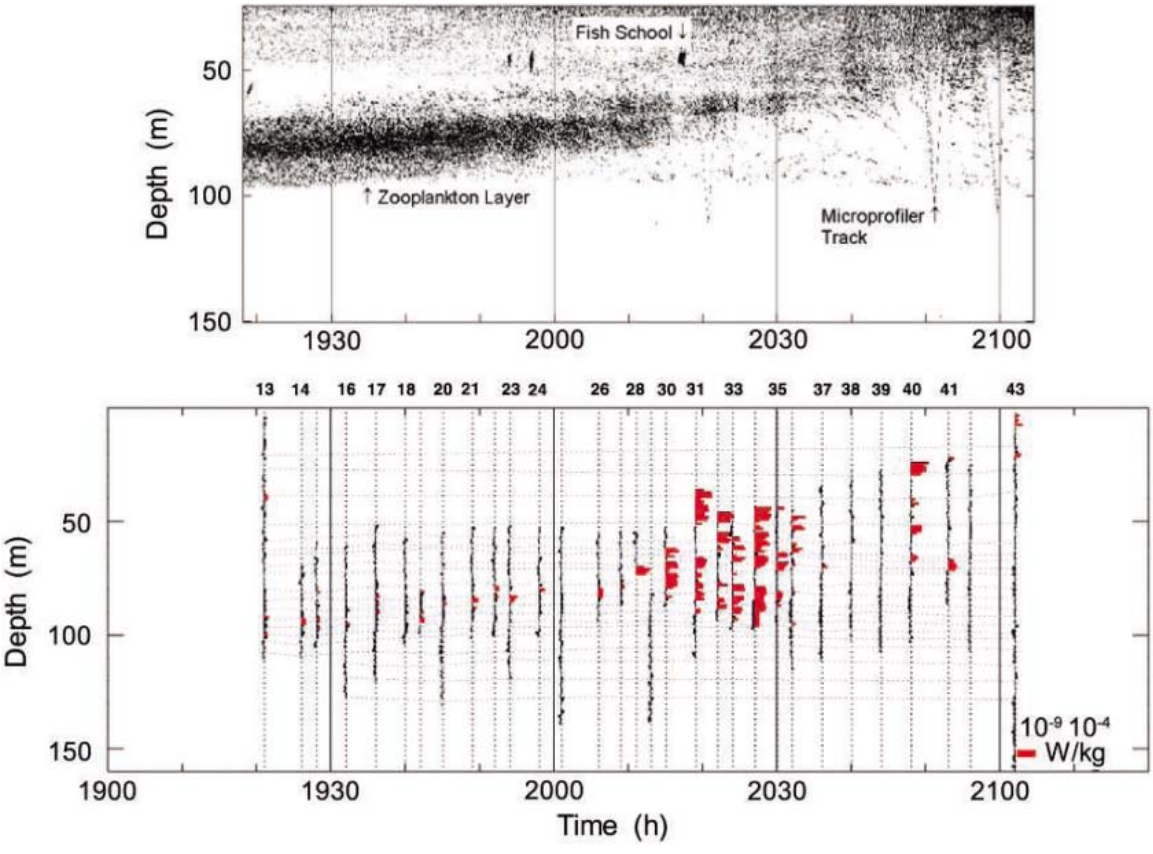

Fig. 1. Profile time series in Saanich Inlet spanning about 100 min during dusk on 28 April 2005. (**Top**) Acoustic backscatter data from a 200-kHz echosounder reveals vertical migration of the backscatter layer. The lowering and raising of the vertical microstructure profiler is also evident for some profiles. (**Bottom**) Turbulent dissipation rate log(ε) (red, index lower right) with vertical dotted lines denoting profile times and horizontal dotted lines denoting salinity or density surfaces. For profiles 13 to 29, dissipation rates are on the order of 10^{-9} W kg^{-1}. For profiles 30 to 36, spanning 17 min, dissipation rates are two to four orders of magnitude higher before falling back to background levels of about 10^{-9} W kg^{-1} for the remainder of the time series.

by previous microstructure measurements. Despite its short duration, this episode was sufficient to boost daily-averaged turbulent eddy diffusivities in Saanich Inlet by two to three orders of magnitude. Daily-averaged diffusivities are 0.02×10^{-4} m^2 s^{-1} when dusk and dawn enhancement are excluded but 4×10^{-4} m^2 s^{-1} to 40×10^{-4} m^2 s^{-1} when they are included.

On a second day of sampling, the sky was overcast. At dusk, the backscattering layer began to migrate, though slightly later than the previous evening. Unlike the first evening, no discernible enhancement of turbulence was observed in the microstructure data. The acoustic data suggest that, for reasons unknown, the lower portion of the backscattering layer (the part most likely composed of the largest euphausiids) remained at depth. We hypothesize that larger euphausiids did not migrate, so that little or no turbulence was generated on the second night.

Several key groups of marine organisms, ranging from krill, to small pelagics such as herring and anchovies, to tuna, occur in sufficient abundance and in sufficiently dense schools to contribute substantial turbulent mixing, particularly in coastal waters (*10*). Our results confirm this for euphausiids in Saanich Inlet. The measured dissipation rates on the order of 10^{-5} W kg^{-1} are consistent with levels predicted for the Antarctic krill *E. superba* (*10*).

These data raise the possibility that a potentially important source of mixing in biologically productive parts of the upper ocean has been overlooked. Further data collected during June 2006 confirm the major results (*25*). Because many densely schooling species, particularly strong vertical migrators, are active near the base of the mixed layer, episodic biologically enhanced turbulence deserves further attention.

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25. Measurements spanning three dusks and two dawns during 10 to 12 June 2006 found no discernible enhancement at the first dusk after an overcast day. After the sunny second and third days, elevated dissipation rates associated with vertical migration of the acoustic backscattering layer were observed during both dusk and dawn.
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Solid Ammonium Sulfate Aerosols as Ice Nuclei: A Pathway for Cirrus Cloud Formation

J. P. D. Abbatt,[1]* S. Benz,[2] D. J. Cziczo,[3] Z. Kanji,[1] U. Lohmann,[3] O. Möhler[2]

Laboratory measurements support a cirrus cloud formation pathway involving heterogeneous ice nucleation by solid ammonium sulfate aerosols. Ice formation occurs at low ice-saturation ratios consistent with the formation of continental cirrus and an interhemispheric asymmetry observed for cloud onset. In a climate model, this mechanism provides a widespread source of ice nuclei and leads to fewer but larger ice crystals as compared with a homogeneous freezing scenario. This reduces both the cloud albedo and the longwave heating by cirrus. With the global ammonia budget dominated by agricultural practices, this pathway might further couple anthropogenic activity to the climate system.

Accurate representation of cirrus clouds remains a challenge to climate modeling, in part because of an incomplete understanding of ice cloud formation mechanisms (*1*). Whereas ice formation studies have been performed at higher temperatures (*2*), only recently has the cold cirrus regime been addressed. This has led to a homogeneous freezing model where ice nucleates directly from the aerosol aqueous phase (*3*). Heterogeneous freezing occurs through selective nucleation onto a small fraction of the background particles (*2*) at lower ice-saturation ratios (S_{ice}) than with homogeneous freezing. Traditionally, it has been thought that good heterogeneous ice nuclei are insoluble solids, such as mineral dust (*2*). Recently, it has been shown in the laboratory that soluble species can also act as ice nuclei in both the immersion (*4*, *5*) and deposition modes (*6*). In the latter case, Shilling *et al.* (*6*) demonstrated that roughly 1 in 10^5 supramicrometer-sized particles of solid ammonium sulfate on a cold plate act as ice nuclei at low ice supersaturations, suggesting that this could be an important atmospheric process. Here, we report measurements of the onset for deposition ice formation on solid ammonium sulfate aerosol under experimental conditions similar to those in the cirrus regime, and we assess the impact of this new ice formation mechanism on past laboratory experiments, field observations, and global climate.

Measurements at Storm Peak, CO, implicate a role of ammoniated particles in selective ice nucleation at low S_{ice} (*7*). It was observed that between 10^{-4} and 10^{-5} of the particles were heterogeneous ice nuclei. Of this fraction, roughly 25% were not classified as conventional insoluble ice nuclei particles—i.e., they did not contain substantial levels of mineral dust, elemental carbon, metal, or fly ash. Instead, these ice nuclei were sulfates, with some degree of organics present. Although the degree of neutralization of the particles was not measured, continental sulfate aerosol likely contains a large amount of ammonium (*8*).

These observations of selective heterogeneous ice nucleation on continental sulfate particles are consistent with laboratory ice nucleation experiments. Figure 1 presents data for two aerosol types representative of sulfate aerosol endpoints. H_2SO_4 particles exist in remote settings away from

[1]Department of Chemistry, University of Toronto, Toronto, ON M5S 3H6, Canada. [2]Institute for Meteorology and Climate Research (IMK-AAF), Forschungszentrum Karlsruhe, Postfach 3640, 76021 Karlsruhe, Germany. [3]Institute for Atmospheric and Climate Science, ETH Zurich, 8092, Zurich, Switzerland.

*To whom correspondence should be addressed. E-mail: jabbatt@chem.utoronto.ca

dominant sources of ammonia such as livestock and nitrogen-based fertilizer (*9*). $(NH_4)_2SO_4$, a major component of continental aerosol, is formed by the uptake of ammonia by H_2SO_4.

Figure 1A shows data for the homogeneous ice formation of H_2SO_4. Figure 1B shows $(NH_4)_2SO_4$ data from recent experiments with solid particles and from older experiments that attempted to measure homogeneous freezing conditions. Because we sought to distinguish the freezing behavior of H_2SO_4 from that of $(NH_4)_2SO_4$, we plotted only the results from homogeneous freezing experiments in which both materials had been studied by the same experimental approach and research group (*10*). Ice formation has been analyzed by means of infrared (IR) interrogation of submicrometer-sized particles in flow tubes (*11*–*14*), optical observation of supermicrometer-sized particles on a hydrophobic support (*15*, *16*), calorimetry of emulsified samples (*16*), optical counting of particles exiting an ice nucleation chamber (*17*), and optical counting/Fourier transform infrared observation in a low-temperature, expansion cloud chamber (*18*, *19*).

H_2SO_4 freezing conditions show good agreement, validating all of the experimental techniques (Fig. 1A). The results match those of the homogeneous freezing model, supporting a homogeneous freezing mechanism for H_2SO_4. By contrast, the freezing conditions for $(NH_4)_2SO_4$ span a wide range (Fig. 1B). The recent experiments for heterogeneous freezing yield low S_{ice} onsets. The literature experiments attempting to measure homogeneous freezing result in a much wider range of freezing conditions.

The relationship between the ice onsets and the fraction of particles freezing provides insight into the wide variability. The experiments most sensitive to a small freezing fraction are the flow tube and cloud chamber (*11*, *13*, *18*, *19*), in which sensitivity is 1 in 10^3 to 10^5 particles—i.e., approximately the ice nuclei fraction exhibited in the field (*2*). This sensitivity arises in the flow tube from a Bergeron-like transfer of water from aqueous particles to a small number of frozen particles (*2*, *20*). By contrast, the hydrophobic support and emulsion experiments (*15*, *16*) measure lower freezing temperatures, in part because they report freezing of a large fraction of the particles and because they study orders of magnitude fewer particles. The results from the ice nucleation chamber indicate that the onset S_{ice} of $(NH_4)_2SO_4$ depends on the aerosol freezing fraction; onsets for 1% freezing were higher than those for 0.1% (Fig. 1B) (*17*).

Why do a select fraction of particles initiate ice formation with $(NH_4)_2SO_4$ but not H_2SO_4? Recent laboratory results indicate that solid $(NH_4)_2SO_4$ particles can act as deposition ice nuclei (Fig. 1B). In particular, under conditions that are similar to those in the atmosphere, we used a cloud chamber to observe ice onsets for both solid and liquid $(NH_4)_2SO_4$ particles. Solid particles that were formed by efflorescence at 295 K and less than 10% relative humidity (RH) nucleate ice efficiently: onset S_{ice} = 1.20 at 221 K. For aqueous particles, the onset is close to that for H_2SO_4: S_{ice} = 1.55 at 219 K. The onset becomes slightly lower and the fraction of deposition ice nuclei increases as aqueous particles are exposed to drier conditions before the ice formation phase of the experiment; that is, additional particles crystallized at the lower RH [supporting online material (SOM) text]. We also studied aqueous $(NH_4)_2SO_4$ particles sprayed onto a hydrophobic support. Ice has also been observed to form selectively at low supersaturations (Fig. 1B). Additional details on the experimental technique and preliminary results at 233 K are described in the SOM text and (*21*, *22*).

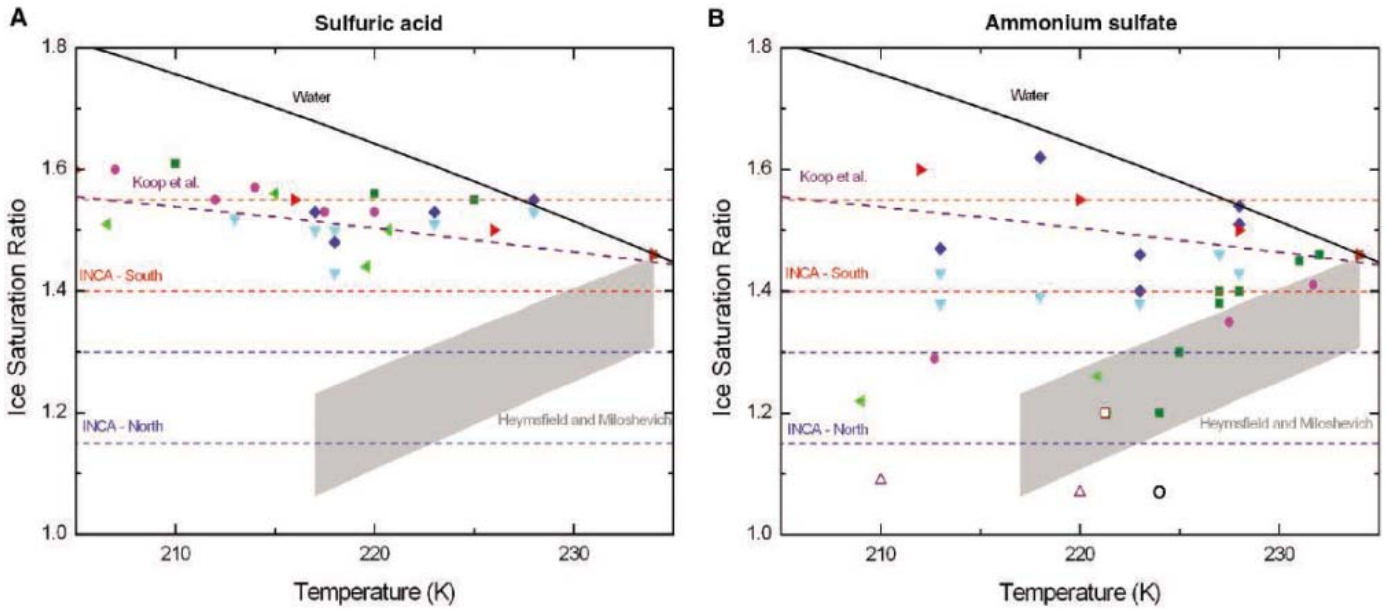

Fig. 1. (**A**) Ice formation points for sulfuric acid, plotted as saturation ratio with respect to ice as a function of temperature. Cziczo and Abbatt (*14*), flow tube/IR (dark green ■); Prenni *et al.* (*12*), flow tube/IR (pink ●); Koop *et al.* (*15*), hydrophobic support/optical (red ▶); Chen *et al.* (*17*), thermal-gradient diffusion chamber, data for 0.1% of the aerosol population freezing (light blue ▼); Chen *et al.* (*17*), thermal-gradient diffusion chamber, data for 1% of the aerosol population freezing (dark blue ◆); Möhler *et al.* (*18*), cloud chamber/IR/optical, data for the first run on each aerosol sample in Experiment Series B, and Mangold *et al.* (*19*), cloud chamber/IR/optical (light green ◀). Also shown are the liquid water saturation line [solid black line (fig. S1)]; water activity–based homogeneous freezing model of Koop *et al.* (*3*) (dashed purple line, nucleation rate = 10^{10} cm^{-3} s^{-1}); bounds for cirrus formation from Heymsfield and Miloshevich (*24*) (shaded gray area); upper and lower limits for cirrus formation in northern INCA campaign (dashed blue lines); and upper and lower limits for cirrus formation in southern INCA campaign (dashed red lines) (*26*). (**B**) Ice formation points for $(NH_4)_2SO_4$, plotted as supersaturation with respect to ice as a function of temperature. Solid symbols show data for homogeneous freezing conditions and open symbols show more recent data for solid particles. Cziczo and Abbatt (*13*), flow tube/IR (dark green ■); Wise *et al.* (*11*), flow tube/IR (pink ●); Bertram *et al.* (*16*), hydrophobic support/optical and emulsion/scanning calorimetry (red ▶); Chen *et al.* (*17*), thermal-gradient diffusion chamber, data for 0.1% of the aerosol population freezing, wet particles with preconditioner (light blue ▼); Chen *et al.* (*17*), thermal-gradient diffusion chamber, data for 1% of the aerosol population freezing (dark blue ◆); and Mangold *et al.* (*19*), cloud chamber/IR/optical, (light green ◀). Also plotted are onsets for heterogeneous ice nucleation by solid $(NH_4)_2SO_4$ as measured in our laboratories with particles deposited on a hydrophobic support (black ○) and in a large cloud chamber (brown □), and by Shilling *et al.* (*6*) (purple △). Additional new data from the cloud chamber are plotted in fig. S1. Solid and dashed lines and gray area are the same as in (**A**).

Deposition ice formation can explain some of the disparity between the published ice onsets in the experiments that sought to study homogeneous freezing of $(NH_4)_2SO_4$. Given that the particles in the flow tubes were sometimes briefly exposed to somewhat reduced RH when mixed into the carrier gas, a fraction may have effloresced at low temperature, thus driving ice nucleation heterogeneously. This finding is also in agreement with new results in which a polycrystalline sample with high surface area was seen to promote selective ice nucleation by deposition nucleation (*6*) but not with the earlier work of Chen *et al*. (*17*) (SOM text). $(NH_4)_2SO_4$ can be an efficient immersion ice nucleus (*4*, *23*). Overall, the findings that soluble species can act as ice nuclei challenge the traditional understanding in cloud physics that only insoluble species can act in this manner (*2*).

Some cirrus cloud formation observations are consistent with ammoniated sulfate particles acting as low-temperature ice nuclei. As noted previously (*14*), cirrus onsets measured in the field correspond with those from the flow tube for both ammonium sulfate and bisulfate. This is shown in Fig. 1B, where bounds for cirrus onset in one continental regime are indicated (*24*). The laboratory measurements sensitive to ice onset of 1 in 10^3 to 10^5 particles, in particular the cloud chamber work that is conducted under the most atmospherically relevant conditions, are similar to the field observations (Fig. 1A). Measurements in the North American free troposphere indicate that particles are neutralized to a large degree by ammonia (*8*). Given that the residence time for such particles can be long, individual particles within the same air volume experience different prior RH and temperatures. As shown by a trajectory modeling study (*25*), it is likely that a large fraction of the total sulfate aerosol will be exposed to RHs sufficiently low to induce efflorescence of solid ammoniated sulfates, particularly in the upper troposphere and Northern Hemisphere.

As a second example, the interhemispheric differences in cirrus properties from anthropogenic emissions (INCA) campaigns studied cirrus formation in clean air of oceanic origin in the Southern Hemisphere (centered at Punta Arenas, Chile, 54°S, March to April) and in moderately polluted air of mixed oceanic-continental origin in the Northern Hemisphere (centered at Prestwick, Scotland, 55°N, September to October). The ice cloud S_{ice} onsets were considerably different between the locations, occurring between 1.15 and 1.30 in the North and between 1.40 and 1.55 in the South (dashed lines in Fig. 1) (*26*). Given that the mean temperature of the measurements was 225 K (*27*), Fig. 1A demonstrates that the conditions for cirrus formation in the South are best matched by those for homogeneous freezing of H_2SO_4, whereas in Fig. 1B there is overlap between northern cirrus formation conditions and those for heterogeneous ice formation by means of $(NH_4)_2SO_4$.

The ammonium-to-sulfate molar ratio of the aerosol was not measured during INCA. However, measurements in the North Atlantic free troposphere indicate ratios that are sometimes close to 2 (*28*). In the Southern Hemisphere, where continental sources of ammonia are less important, the few measurements indicate values of 1 or lower (*29*, *30*). Models predict some difference in the degree of aerosol neutralization between the southern and northern INCA locations at the time of the campaigns (*9*, *31*, *32*). In particular, in the South (March to May), the aerosol is acidic, whereas in the North (September to November), it is more neutralized with compositions between $(NH_4)_2SO_4$ and NH_4NO_3 (*32*). Colberg *et al*. (*25*) predict that a major fraction of the Northern Hemispheric sulfate aerosol will contain solids, whereas liquids will prevail in the Southern Hemisphere.

Haag *et al*. (*27*) have analyzed the INCA data and concluded that a homogeneous freezing mechanism prevails in the South, whereas a coupled mechanism involving homogeneous freezing and selective heterogeneous freezing occurs in the North. However, these authors were not able to identify the chemical nature of the ice nuclei. Based on the laboratory evidence in Fig. 1 and the expected aerosol composition, we believe that cirrus formation occurred by homogeneous freezing of sulfuric acid particles in the southern INCA flights, whereas we raise the possibility that a fraction of ammoniated sulfate particles may have heterogeneously initiated cirrus formation at lower S_{ice} in the North.

As shown by the Storm Peak data, heterogeneous ice nucleation may proceed through a variety of ice nuclei, including mineral dust, metals, soot, solid organics, and, as demonstrated here, $(NH_4)_2SO_4$. Of these, fresh mineral dust is an especially efficient ice nucleus, likely much more efficient than $(NH_4)_2SO_4$ on a per surface area basis. However, there is considerably less mineral dust in the upper troposphere than there is sulfate aerosol, especially away from dust-source regions. Also, the dust can be coated heterogeneously with organics or sulfate, or it may be removed by the initial stages of cirrus formation. Conclusively evaluating the ice nuclei efficiency of $(NH_4)_2SO_4$ and other solid ammoniated sulfates relative to that of dust and other solids, in campaigns such as INCA or in the atmosphere in general, requires measurements of the degree of effloresced sulfate particles and the S_{ice} onset's dependence on particle surface area, mode of preparation (SOM text), and effects of other species present such as organics. In this context, the most atmospherically appropriate measurements we performed are those from the expansion cloud chamber, in which the particles effloresced homogeneously and low total surface areas were used.

To estimate the potential importance of ammoniated ice nuclei, we introduced ammonia into the ECHAM4 General Circulation Model (*22*, *33*). When heterogeneous freezing of either $(NH_4)_2SO_4$ or dust competes with homogeneous freezing, the number of ice crystals is reduced and the crystals grow larger. For $(NH_4)_2SO_4$, this reduces the shortwave cloud forcing (difference between all-sky and clear-sky radiation at the top of the atmosphere) for cirrus clouds between 0.5 and 0.9 W m^{-2}, as can be expected from parcel model studies (*34*) (Table 1). These bigger crystals sediment faster, reducing the ice water content and the longwave cloud forcing between 0.5 and 1.1 W m^{-2} (Table 1). Because the longwave effect partly offsets the shortwave effect, the net effect for $(NH_4)_2SO_4$ is a larger cooling by up to 0.3 W m^{-2} depending on the chosen scenario (SOM text). In the case of dust acting as ice nuclei, the net cooling can amount to 2.5 W m^{-2}, which emphasizes the importance of determining the relative efficiency of these two potential ice nuclei.

If crystalline ammoniated sulfate particles act as ice nuclei, biogeochemical processes that occur in continental regions are connected to ice cloud formation. Atmospheric ammonia has been highly anthropogenically affected, now being released primarily through livestock and nitrogen-based fertilizer, alongside smaller vegetation and oceanic and biomass burning sources (*9*). The impact of these agrarian practices on upper tropospheric ice clouds and climate has not been evaluated.

Table 1. Global annual mean shortwave, longwave, and net cloud forcing (difference between all-sky and clear-sky conditions) at the top of the atmosphere; ice water path; and vertically integrated ice crystal number concentration for the different model simulations. HOM: only homogeneous freezing; DU1 and DU10: heterogeneous freezing whenever the dust ice nuclei concentration exceeds 1 cm^{-3} or 0.1 cm^{-3}, respectively, and homogeneous freezing otherwise; AS1, AS10, and AS100: same categorization as DU1, but instead of dust, 1, 10, or 100% of the $(NH_4)_2SO_4$ concentration, respectively, serve as ice nuclei once the $(NH_4)_2SO_4$ number concentration exceeds 1 cm^{-3}.

	HOM	DU1	DU10	AS1	AS10	AS100
Shortwave cloud forcing (W m^{-2})	−48.2	−48.3	−47.0	−47.7	−47.7	−47.3
Longwave cloud forcing (W m^{-2})	29.5	29.3	25.8	29.0	28.7	28.4
Net cloud forcing (W m^{-2})	−18.7	−19.0	−21.2	−18.7	−19.0	−18.9
Ice water path (g m^{-2})	22.3	21.7	14.1	21.2	20.3	19.4
Ice crystal number (10^6 cm^{-2})	1.01	0.925	0.521	0.789	0.716	0.650

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Global Genetic Change Tracks Global Climate Warming in *Drosophila subobscura*

Joan Balanyá,[1]* Josep M. Oller,[2] Raymond B. Huey,[3] George W. Gilchrist,[4] Luis Serra[1]

Comparisons of recent with historical samples of chromosome inversion frequencies provide opportunities to determine whether genetic change is tracking climate change in natural populations. We determined the magnitude and direction of shifts over time (24 years between samples on average) in chromosome inversion frequencies and in ambient temperature for populations of the fly *Drosophila subobscura* on three continents. In 22 of 26 populations, climates warmed over the intervals, and genotypes characteristic of low latitudes (warm climates) increased in frequency in 21 of those 22 populations. Thus, genetic change in this fly is tracking climate warming and is doing so globally.

Climate change is altering the geographic ranges, abundances, phenologies, and biotic interactions of organisms (*1*, *2*). Climate change may also alter the genetic composition of species, but assessment of such shifts requires genetic data sampled over time (*2*–*5*). For most species, time series of genetic data are nonexistent or rare, especially on continental or global scales (*5*). For a few *Drosophila* species, however, time-series comparisons of chromosomal inversions are feasible (*4*, *6*–*8*) because these adaptive polymorphisms were among the first genetic markers quantified in natural populations (*9*). Consequently, historical records of inversion frequencies in *Drosophila* spp. provide opportunities for evaluating genetic sensitivity to changes in climate and other environmental factors (*4*, *8*, *10*, *11*). Time-series data (13 to 46 years, mean = 24.1 years) of chromosomal-arrangement frequencies and of climate are now available for 26 populations of the cosmopolitan species *D. subobscura* on three continents. Here we examine whether ambient temperatures have warmed at these sites and also whether genotypes characteristic of low latitudes have increased in frequency.

Drosophila subobscura is native to the Old World, where it is geographically widespread from North Africa to Scandinavia (*12*). It has a rich complement of chromosomal arrangements (inversions) on its five acrocentric chromosomes (*12*). Over the past half-century, inversion frequencies have been scored at many sites in the Old World. The frequencies of most inversions change clinally with latitude and thus with climate (*13*, *14*). These climatic clines must be maintained dynamically by natural selection because the gene flow within continents is very high (*15*). Therefore, temporal shifts in inversion frequencies should be sensitive indicators of adaptive responses to climate change (*4*, *10*, *11*).

In the late 1970s, *D. subobscura* was accidentally introduced (*16*) into South America and soon thereafter (*17*) into North America. It spread explosively on both continents (*18*). Geneticists soon (1981 in South America, 1985 to 1986 in North America) began surveying inversion frequencies of these introduced populations at different latitudes (*19*, *20*). On both continents they detected incipient latitudinal clines in chromosome inversion frequencies that almost always had the same sign with latitude as in the Old World, supporting the inference that these clines are adaptive (*18*, *21*). Some other traits of these introduced flies show rapid clinal evolution as well (*22*, *23*).

To obtain comparative data on contemporary chromosome-arrangement frequencies, we and colleagues have revisited many of the historical sampling sites in both the Old and New World. Initial studies with *D. subobscura* reported that "warm-climate" inversions have increased in frequency at several European sites and proposed that these shifts reflect climate warming, but these studies did not investigate continent-scale correlations with climate (*10*, *11*, *24*, *25*). Our analyses here investigate whether the magnitude and direction of genetic shifts actually parallel those in climate, and whether they do so on all three continents.

[1]Department of Genetics, [2]Department of Statistics, Faculty of Biology, University of Barcelona, Diagonal 645, Barcelona 08071, Spain. [3]Department of Biology, Box 351800, University of Washington, Seattle, WA 98195–1800, USA. [4]Department of Biology, Box 8795, College of William and Mary, Williamsburg, VA 23187–8796, USA.

*To whom correspondence should be addressed. E-mail: jbalanya@ub.edu

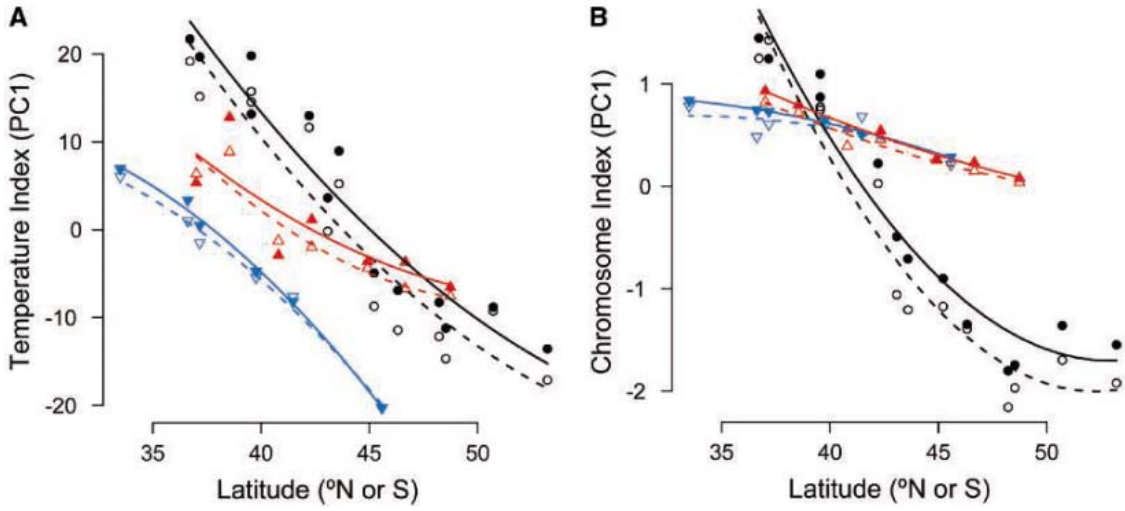

Fig. 1. Temporal shifts in temperature and in chromosome inversion frequencies at different latitudes on three continents. (**A**) A climate temperature index (T_{PC1}) is inversely correlated with latitude for 26 sites on three continents and has increased from the historical (open symbols, dashed regression lines) to contemporary samples (filled symbols, solid regression lines). Black, European sites; red, North American sites; and blue, South American sites. Regression lines are for second-degree orthogonal polynomials. (**B**) A chromosome index (Ch_{PC1}) is inversely related to latitude and has increased from the historical to contemporary samples (see text).

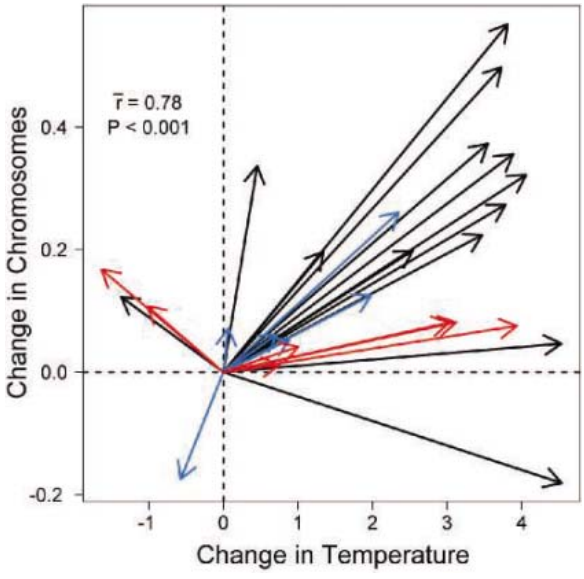

Fig. 2. Change in the direction of the chromosome index over time parallel those in the temperature index at 22 of 26 sites (upper right and lower left quadrants). Black, European sites; red, North American sites; and blue, South American sites.

Table 1. Spearman's rho correlation coefficients (95% confidence limits) for the relation between indices for chromosomes (Ch_{PC1}) and for climate (T_{PC1}) for old and for new samples on three continents. $^{**}P < 0.01$, *** $P < 0.001$.

Sample	Europe	South America	North America
Old	0.94***	0.49	0.93**
	(0.806, 0.982)	(−0.53, 0.930)	(0.584, 0.990)
New	0.95***	1.00***	0.93**
	(0.838, 0.985)	(1, 1)	(0.584, 0.990)

Table 2. Estimated equatorial shift (in degrees of latitude) between old and new samples from 10,000 bootstrapped replications of chromosome clines and of temperature clines. Values show means ± SE, with the 95% confidence limits indicated in parentheses.

Sample	Europe	South America	North America
Chromosomes	−0.884 ± 0.1721	−1.089 ± 1.4785	−0.757 ± 0.2612
	(−1.221, −0.547)	(−3.987, 1.809)	(−1.268, −0.245)
Temperatures	−1.106 ± 0.2095	−0.545 ± 0.1872	−0.735 ± 0.4275
	(−1.516, −0.696)	(−0.912, −0.178)	(−1.573, 0.103)

Historical data on inversion frequencies of *D. subobscura* in the Old and New Worlds were drawn from the literature (*11*). Between 1997 and 2004, contemporary estimates of inversion frequencies were scored from flies at the same (or very nearby) populations (*26*), during the same seasons as the original samples (*11*, *27*). Contemporary samples were also obtained in 2004 for seven populations in North America (*26*) (table S1). In all samples, each of the five acrocentric chromosomes was examined and scored for chromosomal arrangements, according to standard procedures (*26*). We analyzed 50 arrangements, including 21 that show significant latitudinal clines in the Old World and all 18 arrangements present in the New World (*27*).

Rather than analyzing frequency shifts of individual inversions, we developed a genome-wide index based on frequencies (p_i) of all inversions on the five acrocentric chromosomes. Specifically, we applied a principal component analysis to the centered and unscaled frequencies (after transformation by $2\sqrt{p_{ij}}$) of the scored arrangements on all chromosomes for the 52 (population × time) samples (*26*). Here we analyze the first principal component, which accounts for 45.8% of the variance.

To determine whether climates had shifted between samples at the study sites, we developed an index of ambient temperature. We compiled monthly mean temperatures from the nearest recorded weather station for the 4-year period before each sample and then computed a principal component index of the centered, unscaled monthly means for each site and period (*26*). The temperature index (T_{PC1}) reflects overall temperature and accounts for 79.8% of the variation.

T_{PC1} is inversely correlated with latitude on the three continents (Fig. 1A, table S2). Within continents, we found no significant heterogeneity among slopes between temporal samples ($F_{[4,17]} = 0.313$, $P = 0.865$), and so we used analysis of covariance to fit a common slope to

compute the between-sample effect (*28*). T_{PC1} increased significantly between samples ($F_{[1,\ 25]} = 28.8$, $P = 1.22 \times 10^{-6}$), consistent with global climate warming. Indeed, T_{PC1} increased at 22 of 26 sites. Shifts were larger in Europe (Fig. 1A), probably reflecting the longer sample intervals there and the broader range of climates (Fig. 1A).

A genomewide, principal component index of chromosome arrangement frequencies (Ch_{PC1}) was computed for all sites (*26*). Ch_{PC1} is inversely related not only to latitude (Fig. 1B, table S2), but also to T_{PC1} on all three continents (Table 1). Thus, Ch_{PC1} serves as a genetic indicator of the local climate. Because we found no significant differences in slope between temporal samples within continents ($F_{[4,17]} = 1.03$, $P = 0.419$), we fit a common slope within each continent and carried out an analysis of covariance (*29*). If the observed climate warming (Fig. 1A) is having a genetic impact, then genotypes associated with low latitudes (i.e., high Ch_{PC1} scores, Fig. 1B) should have increased in frequency between samples. In 24 of the 26 populations, this was indeed the case ($F_{[1,25]} = 22.7$, $P = 1.99 \times 10^{-6}$) (Fig. 1B). Within-site shifts in the direction of the chromosome index paralleled those of the temperature index in 22 of 26 sites (Fig. 2, sign-test, $P = 5.3 \times 10^{-5}$; Rayleigh test of uniformity, $\bar{r} = 0.78$, $P = 6.8 \times 10^{-8}$). Moreover, chromosome frequencies shifted toward a more low-latitude pattern in 21 of the 22 sites that warmed over the sample interval (upper right quadrant, Fig. 2). Thus, inversion frequencies have changed in step with climate on three continents.

In effect, genotype frequencies and climate at a given site have become more equatorial over the sample intervals (Figs. 1 and 2). Consequently, we rescaled the magnitude of these shifts (*26*) in terms of equivalent degrees of latitude (*4*). For temperature and for genotypes on all three continents, the observed shifts are equivalent to moving the historical sample site ~1° of latitude closer to the equator (Table 2).

Drosophila subobscura is experiencing detectable climate warming on three continents (Fig. 1A). Environmental warming appears to have had a genetic impact on these flies, because frequencies of chromosome inversions associated with warm latitudes have increased in parallel with climate on these continents (Fig. 2). This genetic shift is exceptionally rapid (*25*) and is detectable even for samples separated by fewer than two decades. Genetic shifts paralleling climate warming have been reported recently for a few other insects (*3*, *4*, *8*, *30*), although on more limited geographic scales. In no example to date, however, is it clear whether the observed shifts at given sites reflect local selection, a progressive invasion of genotypes from low latitudes, or both (*11*). Similarly, it is unclear whether the observed genetic changes reflect thermal (*8*, *31*) or seasonal selection (*5*), or correlates thereof.

The increasing numbers of examples documenting genetic (*2–5*, *8*, *10*, *11*), as well as phenotypic (*1*, *2*) responses, to recent climate change are not surprising from an evolutionary perspective, but nonetheless are disturbing from ecological or economic ones, because such changes signal inevitable disruptions in the distributions, population dynamics, and community interactions of organisms (*1*, *2*). Nevertheless, the ability of *D. subobscura* (*10*, *24*, *25*)—and probably other species with short generation times (*3*, *4*, *8*, *32*)—to respond genetically and rapidly to imposed environmental shifts may partially buffer their persistence in a globally warming world (*5*).

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Waking Experience Affects Sleep Need in *Drosophila*

Indrani Ganguly-Fitzgerald,[1]* Jeff Donlea,[2] Paul J. Shaw[2]

Sleep is a vital, evolutionarily conserved phenomenon, whose function is unclear. Although mounting evidence supports a role for sleep in the consolidation of memories, until now, a molecular connection between sleep, plasticity, and memory formation has been difficult to demonstrate. We establish *Drosophila* as a model to investigate this relation and demonstrate that the intensity and/or complexity of prior social experience stably modifies sleep need and architecture. Furthermore, this experience-dependent plasticity in sleep need is subserved by the dopaminergic and adenosine 3′,5′-monophosphate signaling pathways and a particular subset of 17 long-term memory genes.

Sleep is critical for survival, as observed in the human, mouse, and fruit fly (*1–3*), and yet, its function remains unclear. Although studies suggest that sleep may play a role in the processing of information acquired while awake (*4*, *5*), a direct molecular link between waking experience, plasticity, and sleep has not been demonstrated. We have taken advantage of *Drosophila* genetics and the behavioral and physiological similarities between

fruit fly and mammalian sleep (*2*, *3*) to investigate the molecular connection between experience, sleep, and memory.

Drosophila is uniquely suited for exploring the relation between sleep and plasticity. First, fruit flies sleep. This is evidenced by consolidated periods of quiescence associated with reduced responsiveness to external stimuli and homeostatic regulation—the increased need for sleep that follows sleep deprivation (*6*). Second, *Drosophila* has been successfully used to elucidate conserved mechanisms of plasticity. For example, exposure to enriched environments, including the social environment, affects the number of synapses and the size of regions involved in information processing in vertebrates and *Drosophila* (*7*, *8*). In the fruit fly, these structural changes occur in response to experiential information received within a week of emergence from pupal cases (*9*). Although brain plasticity is not limited to this period, the first week of emergence does coincide with the development of complex behaviors in *Drosophila*, including sleep. Hence, daytime sleep, which accounts for about 40% of total sleep in adults, is highest immediately after eclosion and stabilizes to adult levels 4 days after emergence (*3*).

To assess the impact of waking experience during this period of brain and behavioral development, individuals from the wild-type *C-S* strain were exposed to either social enrichment or impoverishment immediately at eclosion and were tested individually for sleep 5 days later (Fig. 1A). Socially enriched individuals (E), exposed to a group of 30 or more males and females (1:1 sex ratio) before being tested, slept significantly more than their socially impoverished (I) siblings, who were housed individually (Fig. 1, B and C; $P < 0.001$). This difference in sleep [ΔSleep (E)] was restricted to daytime sleep. Socially enriched individuals consolidated their daytime sleep into longer bouts of ~60 min compared with their isolated siblings, who slept in 15-min bouts (Fig. 1D, $P < 0.0001$). In contrast, nighttime sleep was unaffected by prior social experience [Fig. 1, B and C; $P = 0.4328$ for 1B by analysis of variance (ANOVA)], corresponding with observations that daytime sleep is more sensitive to sex, age, genotype, and environment, when compared with nighttime sleep (*10*). This effect of social experience on sleep persisted over a period of days (Fig. 1E). Moreover, it was a stable phenotype: When socially enriched, longer-sleeping individuals and socially impoverished, shorter-sleeping siblings were sleep-deprived for 24 hours, they defended their respective predeprivation baseline sleep quotas by returning to these levels after a normal homeostatic response (Fig. 1F and fig. S1).

[1]The Neurosciences Institute, 10640 John Jay Hopkins Drive, San Diego, CA 92101, USA. [2]Washington University in St. Louis, School of Medicine, 660 South Euclid Avenue, Campus Box 8108, St. Louis, MO 63110, USA.

*To whom correspondence should be addressed. E-mail: transposase@gmail.com

Experience-dependent modifications in sleep have long been observed in humans, rats, mice, and cats (*11*–*13*). But what is the nature of the experiential information that modifies sleep need in genetically identical *Drosophila*? Differences in sleep need in socially enriched and socially impoverished individuals were not a function of the space to which they were exposed: Flies reared in 2-cc tubes slept the same as those reared in 40-cc vials (Fig. 1H; $P = 0.5407$). Neither did it arise out of differences in reproductive state or sexual activity between the two groups: Socially impoverished mated and virgin individuals slept the same (Fig. 1I; $P = 0.9450$), as did socially enriched individuals from mixed-sex or single-sex groups (fig. S2). Further, differences in sleep were not a reflection of differences in overall activity (measured as infrared beam breaks) between the two groups (Fig. 1J; $P = 0.6386$). Although social context can reset biological rhythms (*14*), mutations in *clock* (Clk^{jerk}), *timeless* (tim^{01}), and *cycle* (cyc^{01}) disrupt circadian rhythms but had no effect on experience-dependent responses in sleep need (Fig. 1G).

Because social interaction requires sensory input, we next evaluated fly strains that were selectively impaired in vision, olfaction, and hearing. Blind *norpA* homozygotes failed to display a response in sleep to waking experience: Sleep need in *norpA* mutants did not increase after exposure to social enrichment (Fig. 1G; $P = 0.8385$). In contrast, *norpA*/+ heterozygotes with restored visual acuity slept more when previously socially enriched (Fig. 1G; $P < 0.0001$). Attenuating visual signals by rearing wild-type (*C-S*) flies in darkness also abolished the effect of waking experience on sleep (Fig. 1G; $P = 0.7198$). Compromising the sense of smell while retaining visual acuity also blocked experience-dependent changes in sleep need: Socially enriched $smellblind^{1}$ mutants slept the same as their impoverished siblings (Fig. 1G; $P = 0.8478$). As confirmation, we specifically silenced neurons carrying olfactory input to the brain [*Or83b-Gal4*/*UAS-TNT* (*15*)] and observed that sleep in these flies was also not affected by prior waking experience (Fig. 1F; $P = 0.7569$). Auditory cues, however, did not affect the relation between experience and sleep (Fig. 1G; $P < 0.0001$). Finally, sleep need in individual *Drosophila* increased with the size of the social group to which they were previously exposed (Fig. 1K). Socially isolated flies slept the least, whereas those exposed to social groups of 4, 10, 20, 60, and 100 (1:1 sex ratio) showed proportionately increased daytime sleep need (Fig. 1K). When rendered blind, however, flies did not display this relation between sleep need and the intensity of prior social interactions (see *norpA* mutants in Fig. 1K).

If sensory stimulation received during a critical period of juvenile development directs the maturation of the adult sleep homeostat, then subsequent environmental exposure should not affect adult sleep time and consolidation. Alternatively, if experience-dependent modifications in sleep are a reflection of ongoing plastic processes, this phenomenon would persist in the adult. We observed that sleep in flies was modified by their most recent social experience regardless of juvenile experience. Shorter sleeping socially impoverished adults became longer sleepers when exposed to social enrichment (I⇒E) before being assayed (Fig. 2, A to C). Conversely, longer sleeping socially enriched flies became shorter sleepers after exposure to a period of social isolation (E⇒I; Fig. 2, D and E). Moreover, repeated switching of exposure between the two social environments consistently modified sleep, reflecting an individual's most recent experience (fig. S3).

An estimation of neurotransmitter levels in whole brains revealed that short-sleeping, socially impoverished individuals contained one-third as much dopamine as their longer-sleeping, socially stimulated isogenic siblings (Fig. 2F). Silencing or ablating the dopaminergic circuit in the brain [*TH-Gal4/UAS-TNT* and *TH-Gal4/UAS-Rpr* (*16*)] specifically abolished response to social impoverishment in individuals that were reared in social enrichment (Fig. 2H). We obtained similar results when endogenous dopamine levels were aberrantly increased, by disrupting the monoamine catabolic enzyme, arylalkylamine *N*-acetyltransferase, in Dat^{lo} mutants (*17*) (Fig. 2H). Hence, abnormal up- or down-regulation of the dopaminergic system prevented behavioral plasticity in longer sleeping, socially enriched individuals when switched to social impoverishment.

Our observation that dopaminergic transmission affects experience-dependent plasticity in sleep need is particularly compelling, given its role as a modulator of memory (*18*). We thus screened mutations in 49 genes implicated in various stages of learning and memory (*19*–*21*) to assess their impact on experience-dependent changes in sleep need. Of these, only mutations in short- and long-term memory genes affected experience-dependent plasticity in sleep need (Fig. 3). Mutations in *dunce* (dnc^{1}) and *rutabaga* (rut^{2080}) have opposite effects on intracellular levels of adenosine 3′,5′-monophosphate (cAMP), but are both correlated with short-term memory loss. In dnc^{1} mutants, waking experience had no impact on subsequent sleep need (Fig. 3A). This effect was partially rescued in dnc^{1}/+ heterozygotes, but complete rescue was only achieved when a fully functional *dunce* transgene (*22*) was introduced into the null mutant background (Fig. 3A). rut^{2080}, however, selectively abolished the ability of socially enriched adults to demonstrate decreases in sleep after exposure to social impoverishment (Fig. 3A), which was reminiscent of aberrant dopaminergic modulation. Similarly, of the long-term memory genes screened, 17 (~40%) specifically disrupted the change in sleep need in socially enriched adults after exposure to social impoverishment (Fig. 3B). For example, overexpression of the *Drosophila CREB* gene repressor, *dCREB-b*, resulted in socially enriched

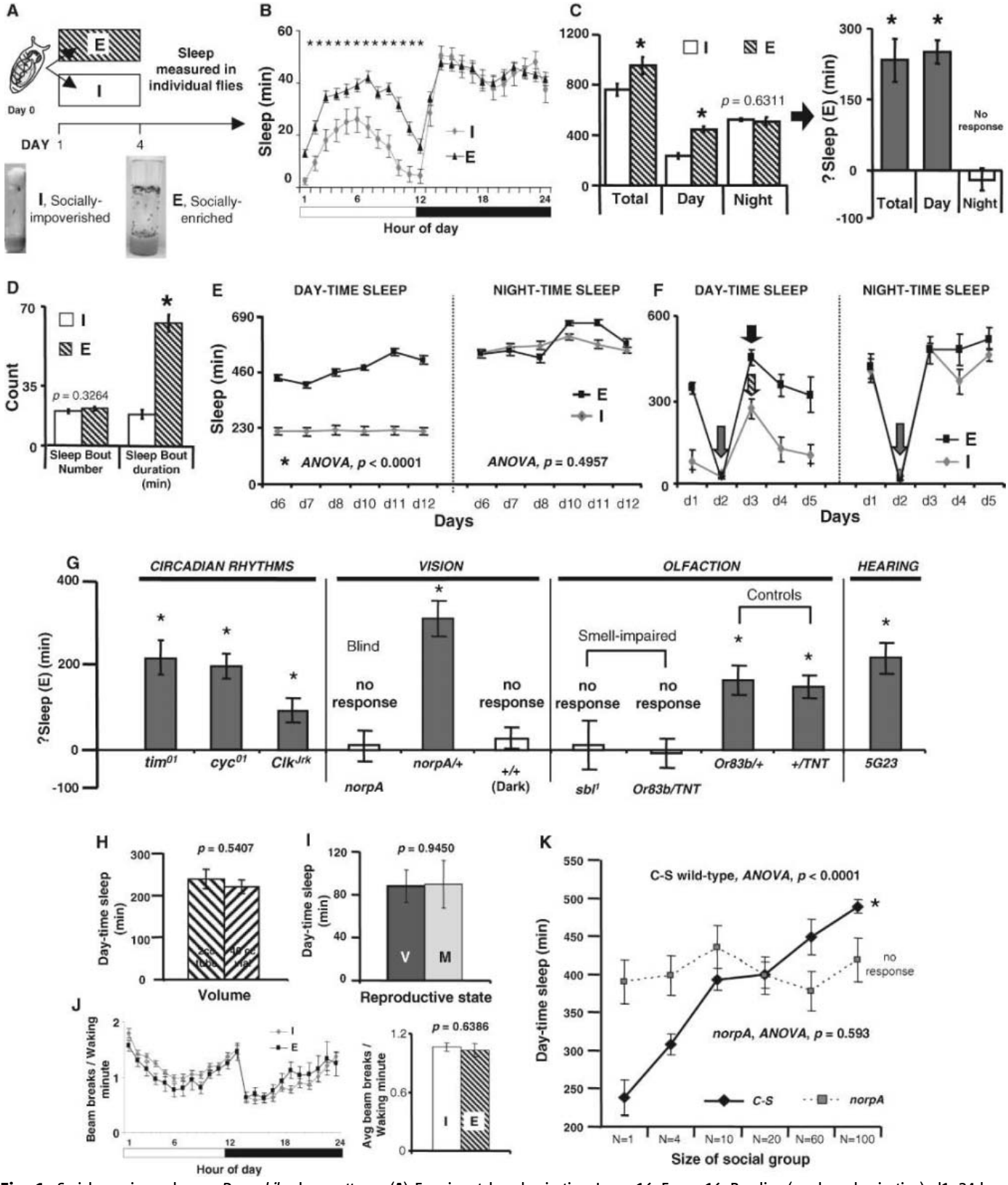

Fig. 1. Social experience changes *Drosophila* sleep patterns. (**A**) Experimental paradigm for juvenile exposure. (**B**) Sleep per hour, over a 24-hour period. (**C**) Total sleep, daytime sleep, nighttime sleep, and ΔSleep: The response in sleep to social enrichment, calculated as a difference in sleep between E individuals and their I siblings. (**D**) Daytime sleep bout number and duration. I, $n = 24$; E, $n = 51$. (**E**) Sleep during 12 days after social exposure. Fruit flies are transferred to fresh food on day 9 (d9). I, $n = 39$; E, $n = 48$. (**F**) Sleep after 24 hours of sleep deprivation. I, $n = 16$; E, $n = 16$. Baseline (no sleep deprivation), d1; 24-hour sleep deprivation, d2; recovery, d3; postrecovery, d4 and d5. (**G**) ΔSleep (E) in circadian, visual, olfactory, and auditory mutants. (**H**) Sleep in fruit flies reared in a 2-cc tube and a 40-cc fly vial. (**I**) Sleep in socially impoverished virgins (V) and mated (M) flies. (**J**) Activity per waking minute each hour over 24 hours. (**K**) Daytime sleep in *C-S* and blind mutants after exposure to increasingly larger social groups. N denotes size of social group. [(A) to (J)] $^*P < 0.005$.

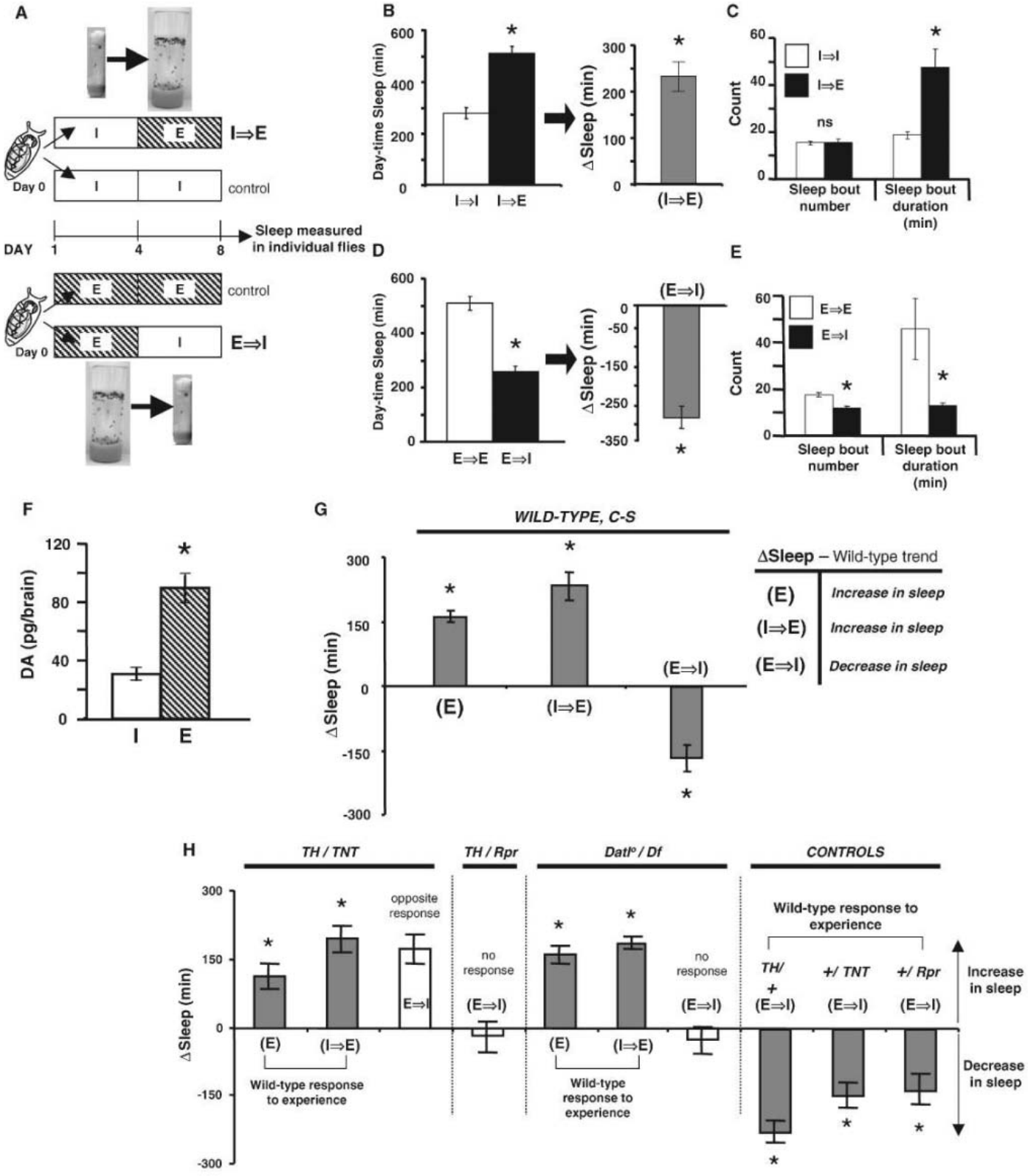

Fig. 2. Plasticity in sleep need and dopamine. (**A**) Experimental paradigm for adult plasticity. (**B** to **E**) Daytime sleep amount, sleep response (ΔSleep), and sleep bout number and duration in I⇒E (n = 20) and E⇒I (n = 55) fruit flies compared with their respective age-matched controls (I⇒I, n = 25; E⇒E, n = 23). (**F**) Dopamine content in whole brains. (**G**) ΔSleep in *C-S* flies. (**H**) ΔSleep in strains with aberrant dopaminergic transmission. In the case of E⇒I *TH/TNT*, flies show an aberrant increase in sleep. *P < 0.005.

flies that continued to be longer sleepers even after exposure to social impoverishment (Fig. 3B). As a control, overexpression of the *dCREB-a* activator yielded wild-type phenotypic read out (Fig. 3B). It is noteworthy that not all long-term memory mutants had a disrupted relation between experience and sleep. Instead, the particular subset of genes identified, only half of which are expressed in the mushroom bodies (*21*), may specifically contribute to pathways that underlie sleep-dependent consolidation of memories.

Finally, to assess the correlation between sleep and memory, male flies trained for a courtship conditioning task that generated long-term memories were measured for sleep after training. Males whose courtship attempts are thwarted by nonreceptive, recently mated females or by males expressing aphrodisiac pheromones form long-term associative memories as evidenced by subsequently reduced courtship of a receptive virgin female (*23*, *24*). Trained males that formed long-term memories slept significantly more than their untrained siblings and wake controls (ones that were sleep-deprived while the experimental flies were being trained; Fig. 4, A to D). Exposure to a virgin female did not alter sleep need. As before, this increase in sleep was associated with longer daytime sleep bouts in trained individuals compared with controls (Fig. 4C). Further, sleep deprivation for 4 hours immediately after training abolished training-induced changes in sleep-bout duration (24 ± 4 min in trained versus 18 ± 3 min in naïve controls, $P = 0.3617$), as well as courtship memory (Fig. 4, A and E). Although these results are intriguing, invertebrate memory is particularly sensitive to extinction by mechanical perturbations. However, gentle handling that ensured wakefulness, but not mechanical stimulation, immediately following training, also abolished subsequent courtship memory (fig. S4). Furthermore, sleep deprivation per se did not affect the formation of long-term memory: Trained flies that were allowed to sleep unperturbed for 24 hours and then subjected to 4 hours of sleep deprivation retained courtship memory (Fig. 4, A and E).

In summary, we demonstrate a rapid and dynamic relation between prior social experience and sleep need in a genetically tractable model organism, *Drosophila melanogaster*. In particular, we report that experience-dependent changes in sleep need require dopaminergic modulation,

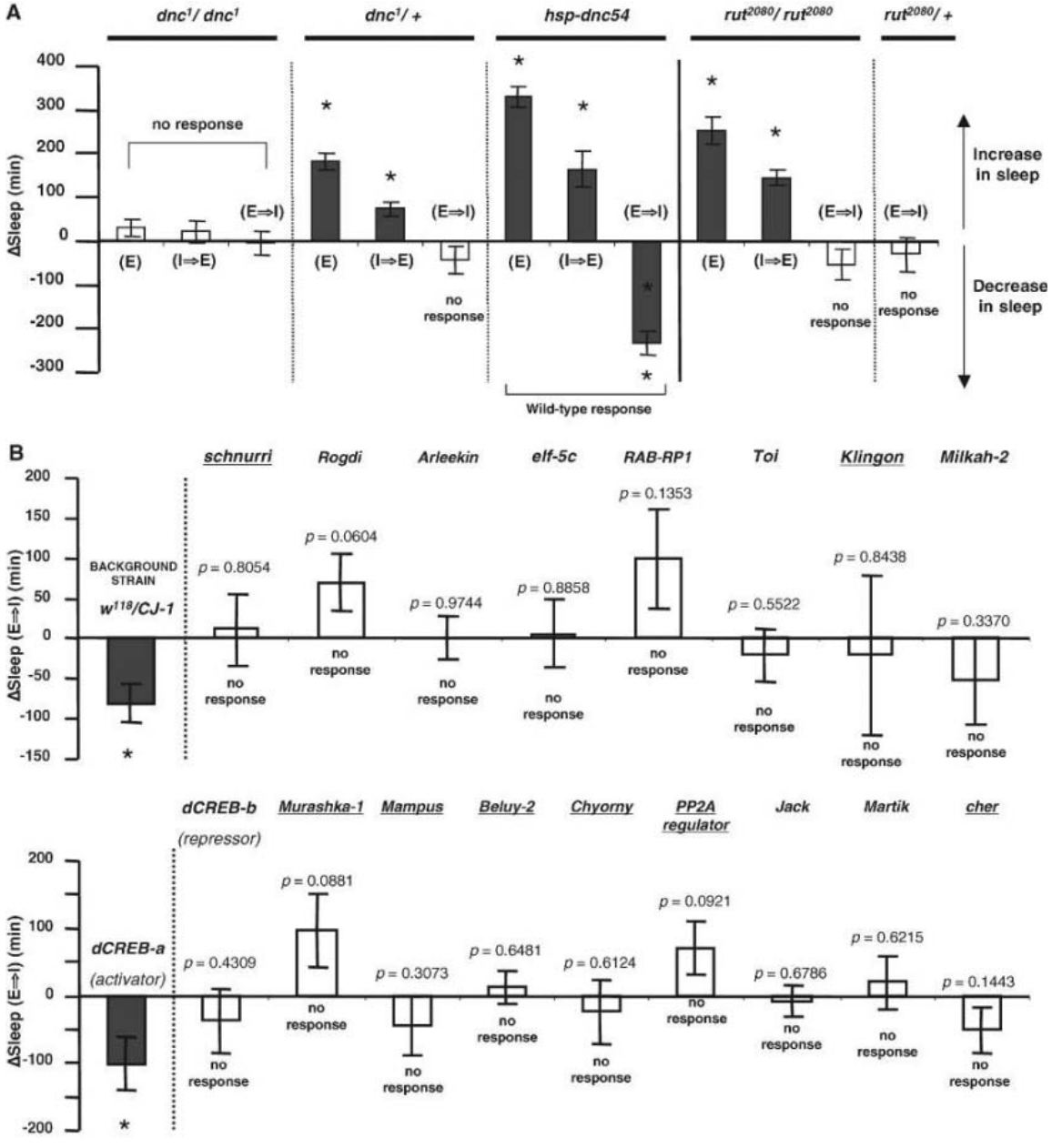

Fig. 3. Long- and short-term memory mutants are resistant to experience-dependent changes in sleep. (**A**) ΔSleep in short-term memory mutants, *dnc*^{1}, *rut*^{2080}, and controls. (**B**) ΔSleep (E⇒I) in *w*^{118}*/CJ-1* wild-type background strain, *dCREB-a* (memory activator) and *dCREB-b* (memory repressor) heat-inducible strains, and 17 of 43 long-term memory mutants that demonstrated disrupted experience-dependent changes in sleep. Underlined genes are not expressed in the mushroom bodies. *P < 0.001.

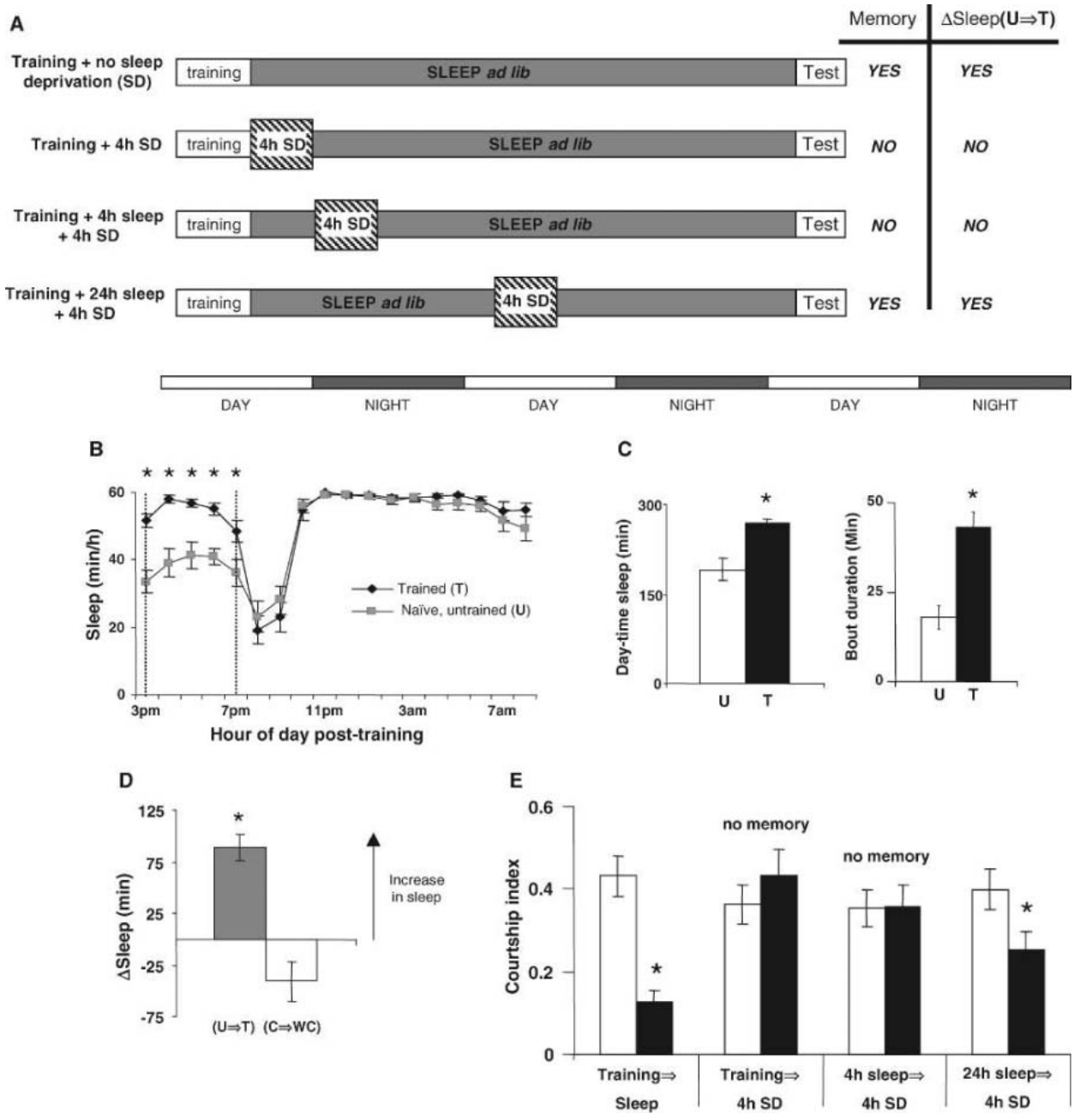

Fig. 4. Formation of associative memories is correlated with posttraining increases in sleep. (**A**) Schematic of experimental design. (**B**) Sleep following training for courtship conditioning in trained (T) and untrained (U) males. (**C**) Daytime sleep amount and bout duration. (**D**) ΔSleep in trained and untrained flies [ΔSleep (U⇒T)] compared with ΔSleep in untrained wake controls (WC) and unperturbed controls (C) [ΔSleep (WC⇒C)]. (**E**) Courtship index (ratio of the percentage of time spent courting to total time of exposure) in T and U flies, after training and following sleep deprivation (SD).

cAMP signaling, and a particular subset of long-term memory genes—supporting the hypothesis that sleep and neuronal activity may be inexorably intertwined. These observations are compelling given two recent studies (*25*, *26*) demonstrating a central role of the mushroom bodies in sleep regulation and emphasize the importance of establishing *Drosophila* as a model system to investigate the molecular pathways underlying sleep and plasticity.

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www.sciencemag.org/cgi/content/full/313/5794/1775/DC1
Materials and Methods
Figs. S1 to S4
References and Notes

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Exogenous Induction of Cerebral β-Amyloidogenesis Is Governed by Agent and Host

Melanie Meyer-Luehmann,[1*] Janaky Coomaraswamy,[1*] Tristan Bolmont,[1,2*] Stephan Kaeser,[1] Claudia Schaefer,[1] Ellen Kilger,[1] Anton Neuenschwander,[3] Dorothee Abramowski,[3] Peter Frey,[3] Anneliese L. Jaton,[3] Jean-Marie Vigouret,[3] Paolo Paganetti,[3] Dominic M. Walsh,[4] Paul M. Mathews,[5] Jorge Ghiso,[6] Matthias Staufenbiel,[3] Lary C. Walker,[7]† Mathias Jucker[1,2]†

Protein aggregation is an established pathogenic mechanism in Alzheimer's disease, but little is known about the initiation of this process in vivo. Intracerebral injection of dilute, amyloid-β (Aβ)–containing brain extracts from humans with Alzheimer's disease or β-amyloid precursor protein (APP) transgenic mice induced cerebral β-amyloidosis and associated pathology in APP transgenic mice in a time- and concentration-dependent manner. The seeding activity of brain extracts was reduced or abolished by Aβ immunodepletion, protein denaturation, or by Aβ immunization of the host. The phenotype of the exogenously induced amyloidosis depended on both the host and the source of the agent, suggesting the existence of polymorphic Aβ strains with varying biological activities reminiscent of prion strains.

The accumulation of misfolded proteins is a common feature of several neurodegenerative disorders. In Alzheimer's disease (AD), the multimerization of the Aβ peptide is an early and central process in the pathogenic cascade (*1*–*3*), but little is known about the mechanisms that govern the initiation of Aβ aggregation and deposition in vivo. Ordered protein aggregation in vitro is a function of protein concentration and time and follows a crystallization-like polymerization mechanism that can be rapidly initiated by introducing an exogenous seed (*4*). In vivo, seeded aggregation of Aβ is seen after injecting AD brain extracts into the brains of non-human primates (*5*) or APP-transgenic mice (*6*), reminiscent of the conformational conversion mechanism of prion infectivity (*7*–*9*).

We injected 10% (w/v) extracts of brain homogenates from autopsied AD patients (AD extract) or from aged, β-amyloid–laden APP23 transgenic mice (*10*) (APP23 Tg extract) into the hippocampus of young male APP23 mice. Four months later, the host mice were analyzed (*11*). Both AD extract and APP23 Tg extract induced robust deposition of Aβ in the hippocampus (Fig. 1, A and B). Intracerebral injection of tissue extract from an aged control patient induced only minimal Aβ deposits, consistent with the low Aβ load in the donor (Fig. 1C; fig S1). No seeded Aβ deposits were found after control injections of brain extract from an aged, wild-type mouse or of phosphate-buffered saline (PBS) (Fig. 1, D and E). Infusion of APP23 Tg extract into wild-type mice did not induce Aβ deposition; thus, the observed Aβ deposits did not simply represent the

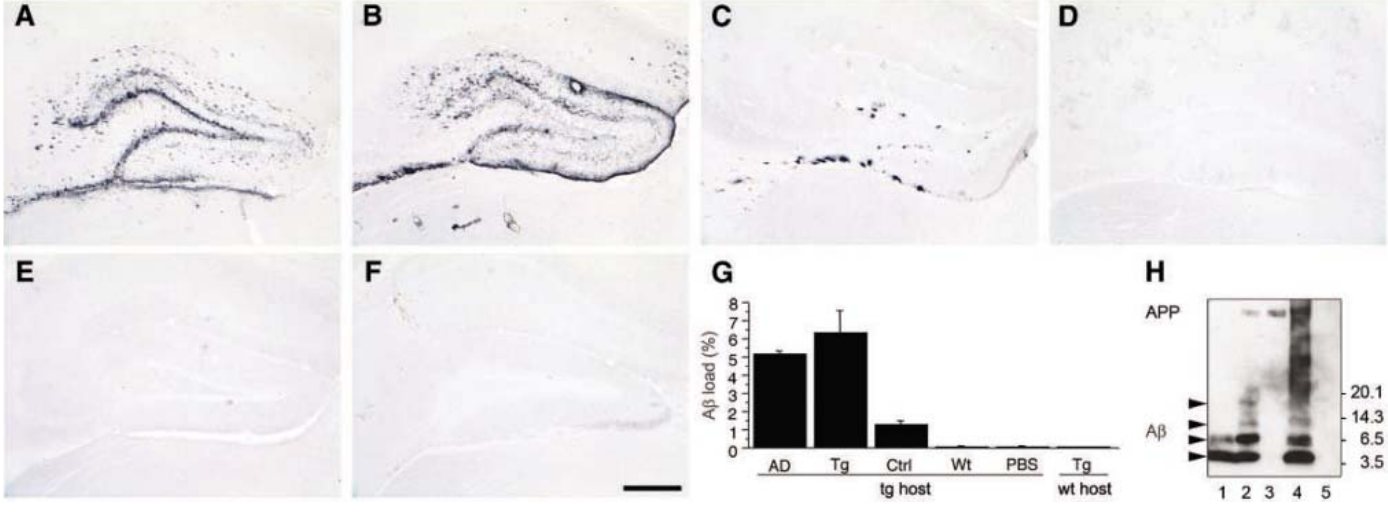

Fig. 1. Brain extract (10%) was injected into the hippocampus of 5-month-old male APP23 hosts (**A** to **E**) and nontransgenic littermates (**F**). Mice were analyzed 4 months later. Injection of AD extract (A) and APP23 Tg brain extract (B) induced numerous Aβ-immunoreactive deposits. Few or no Aβ deposits were detected after injections of brain extract from an aged (95 years) control (Ctrl) patient (C) or wild-type (Wt) mouse (D). No Aβ deposits were observed after PBS injections (E) or when Tg extract was injected into wild-type mice (F). Stereological quantification of Aβ load by immunohistochemistry (**G**) confirmed significant amyloid induction by AD and Tg brain extracts compared to Ctrl and Wt extracts (n = 5 mice per group; mean ± SEM, $P < 0.001$). (**H**) Tris-Tricine SDS–polyacrylamide gel electrophoresis (PAGE) followed by immunoblotting with a human Aβ-specific antibody. Lane 1: synthetic Aβ1-40 + Aβ1-42 (1 ng/μl each); lane 2: AD brain extract; lane 3: Ctrl brain extract; lane 4: APP23 Tg brain extract; and lane 5: Wt brain extract. For each extract, 1 μl was loaded. Arrowheads indicate monomeric, dimeric, trimeric, and tetrameric Aβ. Aβ concentration in the control patient extract was below the detection level. When less dilute samples were used, Aβ was detected in the Ctrl extract, consistent with the sparse amyloid plaques in this patient (fig. S1) and the modest seeding activity of the Ctrl extract. Scale bar, 350 μm.

injected Aβ-containing material (Fig. 1F), and imply that host factors are critical for in vivo seeding. No seeded Aβ deposits were observed when APP23 Tg extract from a young, 2-month-old, predepositing mouse was injected into APP23 hosts. The concentrations of Aβ in the AD and APP23 Tg extracts were estimated to be 1 to 10 ng/μl. In both AD extract and APP23 Tg extract, Aβ monomers, oligomers, and larger multimeric species were present (Fig. 1H).

The localization and biochemical nature of the induced Aβ lesions were markedly similar to those seen in normal, aged APP23 transgenic mice (*10*, *12*). Induced Aβ deposits were found primarily in the injected hippocampus, but some were also observed in the dorsal lateral geniculate nucleus, corpus callosum, and entorhinal cortex, and in the vasculature of the thalamus and pia mater (fig. S2). Most induced Aβ deposits were diffuse, although some congophilic amyloid plaques were present and surrounded by activated microglia, astrocytes, and dystrophic neurites (fig. S2). Aβ in micropunches taken from the hippocampus of injected APP23 mice was mainly parechymal and consisted of Aβ1-40 and Aβ1-42, whereas the amyloid in the thalamic micropunches was mostly vascular and consisted predominantly of Aβ1-40 (fig. S3).

The exogenous induction of Aβ deposition in vivo also is time and concentration dependent (fig. S4). Immunoreactive Aβ deposits first appeared 2 months after injection and thereafter increased significantly with time. APP23 extract that was further diluted to 0.5% produced a pattern of deposition in the host similar to that seen with the 10% extract, but with much less potency (fig. S4).

The phenotype of exogenously induced β-amyloid deposits is dependent on both the agent and the host. We injected brain extract from aged APP23 mice into APP–presenilin-1 (PS1) transgenic hosts, and vice versa (Fig. 2). Amyloid deposition in the hippocampus of male APP23 mice normally begins at 9 to 10 months of age, whereas APPPS1 mice develop Aβ deposition in the hippocampus at 3 to 4 months of age (*10*, *13*). Total Aβ concentration was similar in the two extracts (Fig. 2I), but in APPPS1 extracts, the highly amyloidogenic Aβ1-42 was several times as abundant as Aβ1-40, whereas in extracts from

[1]Department of Cellular Neurology, Hertie-Institute for Clinical Brain Research, University of Tübingen, D-72076 Tübingen, Germany. [2]Institute of Pathology, University of Basel, CH-4003 Basel, Switzerland. [3]Novartis Institutes for Biomedical Research, CH-4002 Basel, Switzerland. [4]Conway Institute of Biomolecular and Biomedical Research, University College Dublin, Dublin, Republic of Ireland. [5]Nathan Kline Institute, New York University School of Medicine, Orangeburg, NY 10962, USA. [6]Departments of Pathology and Psychiatry, New York University School of Medicine, New York, NY 10016, USA. [7]Yerkes National Primate Research Center and Department of Neurology, Emory University, Atlanta, GA 30322, USA.

*These authors contributed equally to this work.
†To whom correspondence should be addressed. E-mail: mathias.jucker@uni-tuebingen.de (M.J.); lary.walker@emory.edu (L.C.W.)

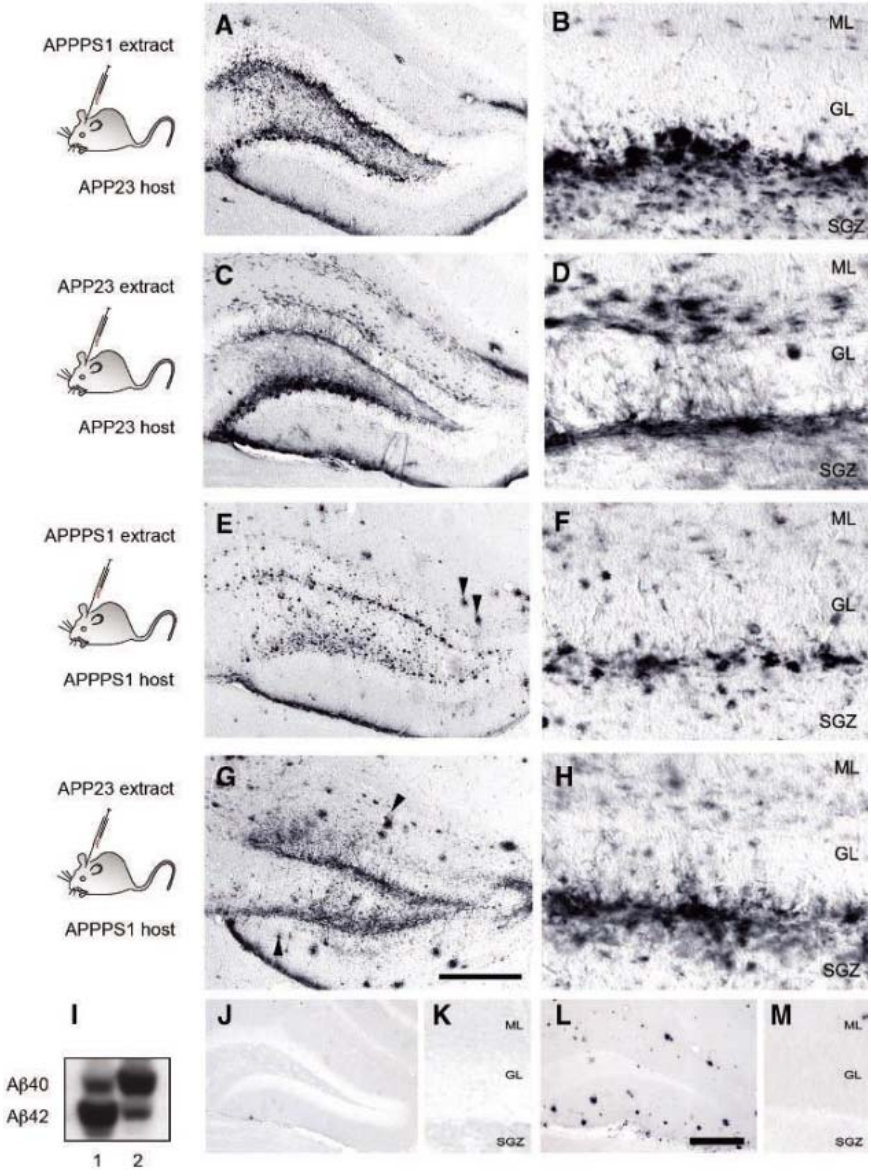

Fig. 2. Brain extract (10%) from an aged APPPS1 mouse or an aged APP23 mouse was injected intrahippocampally into either 5-month-old male APP23 or 2-month-old male APPPS1 hosts. Brains were immunohistochemically analyzed for Aβ 3 months later. The panels to the left (**A**, **C**, **E**, and **G**) show overviews of the hippocampus, and the panels to the right (**B**, **D**, **F**, and **H**) show corresponding higher magnification images of the upper blade of the dentate gyrus (ML, molecular layer; GL, granular cell layer; SGZ, subgranular cell layer). The coarse, punctate pattern of Aβ staining in the APP23 host is apparent, particularly in the SGZ, after injection of the APPPS1 extract (A and B). Injection of APP23 extract induced more filamentous and diffuse Aβ in the ML (C and D). When APPPS1 extract was injected into the APPPS1 host, the coarse, punctate pattern of Aβ induction was even more distinct (E and F), whereas the APP23 extract in APPPS1 hosts induced Aβ lesions intermediate in appearance to the coarse and filamentous types (G and H). At 5 months of age, APPPS1 hosts had developed some Aβ deposits endogenously, predominantly in the ML [arrowheads in (E) and (G)]. For comparison, an 8-month-old noninjected male APP23 mouse (**J** and **K**) and a 5-month-old noninjected male APPPS1 mouse (**L** and **M**) are shown. Scale bar, 350 μm. (**I**) Urea-based SDS-PAGE immunoblot analysis with an antibody specific to human Aβ. Lane 1: APPPS1 extract; lane 2: APP23 extract. Aβ1-42 was the major Aβ species in APPPS1 mice, whereas Aβ1-40 predominated in the APP23 extract. Total Aβ was comparable in the two extracts [APPPS1: total Aβ, 14.7 ng/μl (Aβ40: 4.6 ng/μl; Aβ42: 10.1 ng/μl); and APP23: total Aβ, 11.7 ng/μl (Aβ40: 8.1 ng/μl; Aβ42: 3.6 ng/μl)].

APP23 mice, more Aβ1-40 was present. The amyloid-inducing activity of APP23 and APPPS1 extracts was quantitatively similar 3 months after infusion, but injection of APPPS1 extract into APP23 hosts consistently induced a coarse pattern of compact, punctate Aβ deposition that was mainly confined to the subgranular layer of the hippocampus, whereas the APP23 extract injected into APP23 hosts yielded primarily diffuse and filamentous lesions, with substantial diffuse Aβ in the molecular layer (Fig. 2, A to D). When the same extracts were injected into the APPPS1 host, lesions induced by the APPPS1 extract were even more coarse and punctate, whereas those induced by the APP23 extract were a mixture of filamentous and compact types (Fig. 2, E to H). In mice analyzed 1 month after the infusion of the extract, appreciable coarse Aβ induction was already apparent in APPPS1 hosts but not in APP23 hosts.

To establish whether Aβ is a prerequisite for the amyloid-inducing activity of the extract, APP23 Tg extracts were either Aβ-immunodepleted (Fig. 3, A to C) or mixed with the Aβ-specific antibody (anti-Aβ) Beta-1 (Fig. 3, D to F) and injected into young APP23 hosts. Immunodepletion completely prevented the amyloid-inducing activity of the extract, whereas the Beta-1–containing extract attenuated amyloid induction by >60% compared to a control antibody.

Passive immunization of APP23 host mice with Beta-1 antibodies inhibited the development of induced lesions. Administration of the antibodies commenced 4 weeks after the intracerebral injection of APP23 Tg extract in order not to interfere with the initial seeding process. Serum anti-Aβ titers of 1:2400 to 1:8000 were maintained until mice were killed 4 months after the extract infusion. Amyloid induction was almost completely inhibited in immunized mice compared to those injected with control antibody

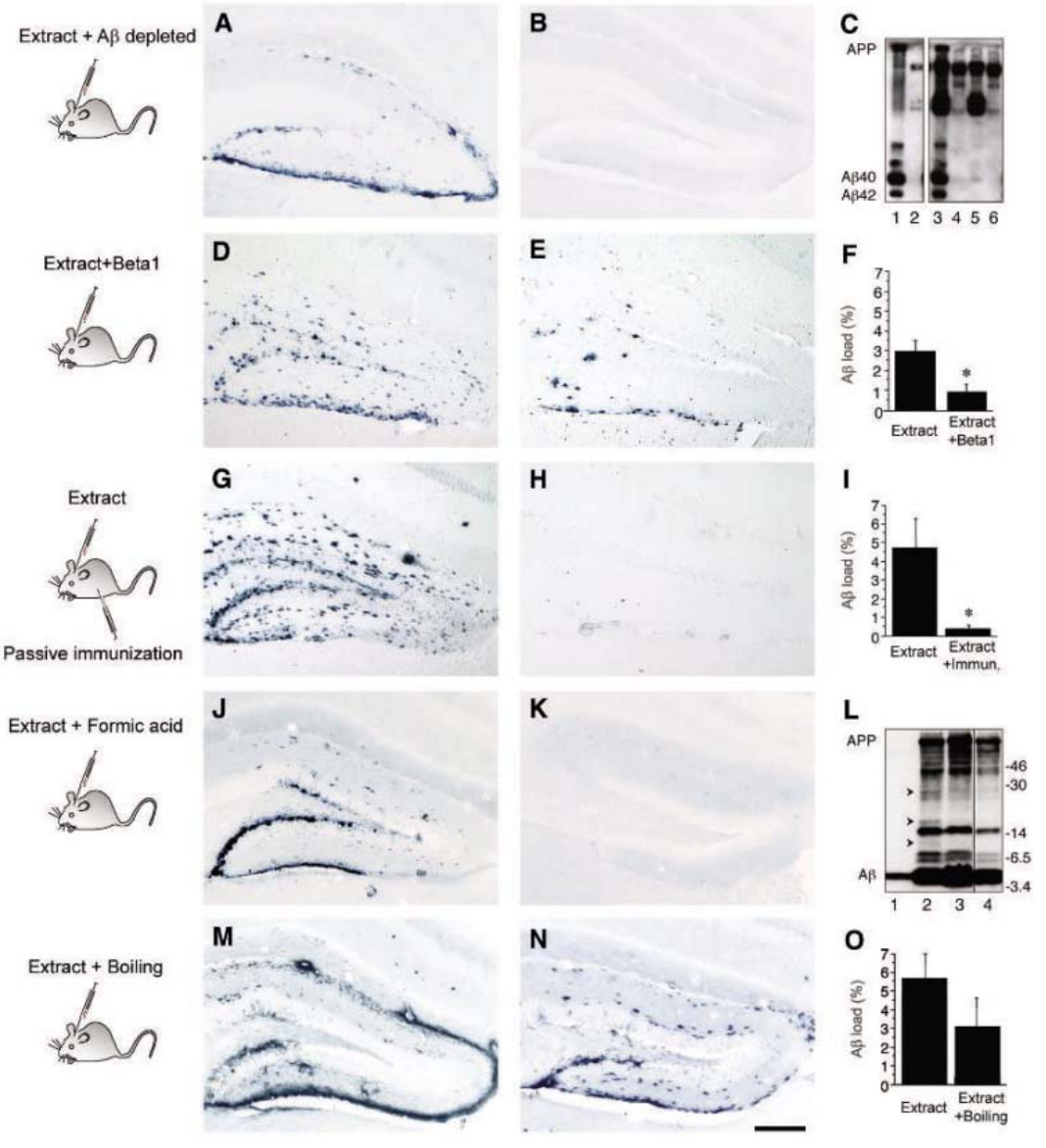

Fig. 3. (**A** to **C**) Immunodepletion: APP23 Tg brain (0.5%) extract (A) or the same extract Aβ-immunodepleted (B) was injected intrahippocampally into 3-month-old APP23 mice that were then analyzed 3 months later. Immunodepletion completely eliminated the amyloid-inducing activity of the extract. (C) Urea-based SDS-PAGE followed by immunoblotting with human Aβ-specific antibody. Lane 1: intact APP23 Tg extract; lane 2: immunodepleted extract (the absence of detectable Aβ and APP fragments is apparent); lanes 3 to 6: eluted pellet fractions after the first, second, third, and fourth depletion steps. (**D** to **F**) Antibody blocking: APP23 or AD brain extract (10%) was mixed with either a control antibody or with anti-Aβ Beta-1 and injected into 6-month-old male APP23 mice. Four months after injection, amyloid induction was significantly lower in the Beta-1–injected mice (E) than in the control mice (D), as confirmed by stereological quantification of immunoreactive Aβ (F) ($n = 5$ mice per group; $P < 0.05$). (**G** to **I**) Immunization: APP23 or AD brain extract (10%) was injected intrahippocampally into 6-month-old male APP23 mice, followed by weekly peripheral injections of Beta-1 antibody (H) or control antibody (G), starting 1 month after the brain-extract injections. Four months later, immunohistochemical (G and H) and stereological (I) analysis revealed a >90% inhibition of amyloid induction by passive immunization ($n = 5$ mice per group; $P < 0.001$). (**J** to **L**) Formic acid treatment: APP23 Tg extract (10%) was treated with formic acid and injected into the hippocampus of 3-month-old APP23 mice that were analyzed 3 months later. (K) Formic acid completely nullified the amyloid-inducing activity of the extract. (J) PBS-treated control extract. (L) Bicine-Tris SDS-PAGE (without urea) followed by immunoblot analysis with human Aβ-specific antibodies. Lane 1: synthetic Aβ (2 ng/μl); lane 2: PBS-treated control APP23 extract (10 μl); and lanes 3 and 4: formic acid–treated APP23 extract (20 μl in lane 3 and 10 μl in lane 4). The oligomeric bands are absent or greatly reduced (arrowheads) in the formic acid–treated extract, even at twice the concentration of the control extract. (**M** to **O**) Heating: APP23 extract (10%) was heated to 95°C (N) before injection into the hippocampus of 3-month-old APP23 mice. Analysis 3 months later showed that heating reduced (45%) but did not eliminate Aβ seeding ($n = 3$ mice per group; $P > 0.05$). Scale bar, 350 μm.

(Fig. 3, G to I). An active immunization protocol revealed similar inhibition (fig. S5).

The amyloid-inducing activity of the extract was disrupted by formic acid denaturation. APP23 Tg extracts were treated for 1 hour with 70% formic acid, followed by dialysis, or heated to 95°C for 5 min, cooled, and injected into young APP23 mice. Formic acid treatment completely abolished the amyloid-inducing activity of the extract, whereas heating reduced, but did not eliminate, amyloid induction (Fig. 3, J to O).

Preparations of soluble or fibrillar synthetic Aβ40, Aβ42, or a mixture of both, in amounts similar to those of Aβ in APP23 Tg brain extracts, failed to induce detectable Aβ deposition (Fig. 4). Injection of synthetic Aβ at concentrations 100 to 1000 times that of Aβ in APP23 Tg brain extracts resulted in amorphous masses of material near the injection site that consisted primarily of the injected material. Few newly generated aggregates were present, as confirmed by infusion of biotinylated Aβ1-42 (fig. S6). Synthetic Aβ oligomers (*14*–*16*) also were ineffective at inducing parenchymal and vascular amyloid, as was Aβ isolated from the conditioned media of cells stably transfected with APP (table S1; fig. S6). Addition of cofactors such as ApoE4 and Cu/Zn, which are thought to promote the polymerization of Aβ (*17*, *18*), did not augment the potency of synthetic material (table S1). Injection of synthetic Aβ mixed with brain extract from wild-type mice also did not produce notable β-amyloidosis (table S1). Finally, the possibility that poor seeding by synthetic Aβ results from the activation of the Aβ-degrading enzyme neprilysin (*19*) or a humoral immune response (*20*) was ruled out by unchanged neprilysin immunoreactivity and by the absence of serum anti-Aβ antibodies in mice injected with synthetic Aβ.

Thus, cerebral extracts induce Aβ deposition in vivo by supplementing (or anticipating) endogenously generated Aβ seeds with exogenous seeds that probably consist of a form of multimeric Aβ. The host-specific morphology and distribution of the induced lesions underscore the essential role of the host in regulating pathogenesis, but the inducing agent also contributes to the pathologic phenotype. The inhibition of seeding by specific immunoneutralization of Aβ (*20*) or formic acid denaturation of the extracts (*21*) suggests that the active agent consists of an aggregated Aβ species (*20, 21*). The finding that synthetic Aβ lacks amyloid-inducing activity in vivo was not unexpected, inasmuch as prion disease has also been difficult to transmit by in vitro–generated (recombinant) prions (*8*).

Synthetic and cell culture–derived Aβ, in concentrations similar to those tested in the present study, are neurotoxic in vivo (*14*, *22*, *23*) and can impair long-term potentiation and cognitive function (*24*, *25*). These observations, in light of the highly variable seeding efficacy of in vitro and in vivo preparations, suggest the occurrence of various Aβ conformations with partially distinct biological activities (*26*), similar to prions (*27*, *28*). Polymorphic and self-propagating synthetic Aβ strains recently have been reported (*9*). Thus, Aβ multimers in vivo also may be polymorphic and polyfunctional, again reminiscent of prions, in which infectivity is strain-dependent and fully encoded in distinct multidimensional conformations (*29*). Whether oligomeric forms of Aβ that are thought to be key cytoactive disease agents (*25*, *30*, *31*) can also be seeded in vivo remains to be determined.

There is currently no evidence that β-amyloidosis (and in particular AD) is transmissible in the same sense as are prion diseases, which can be transmitted to wild-type hosts via diverse routes of varying efficiency and involve systemic cellular mechanisms of prion uptake and distribution (*7*, *32*). However, an understanding of the mechanisms involved in the instigation and propagation of abnormal Aβ assemblies in vivo could shed light on the origins of idiopathic Alzheimer's disease.

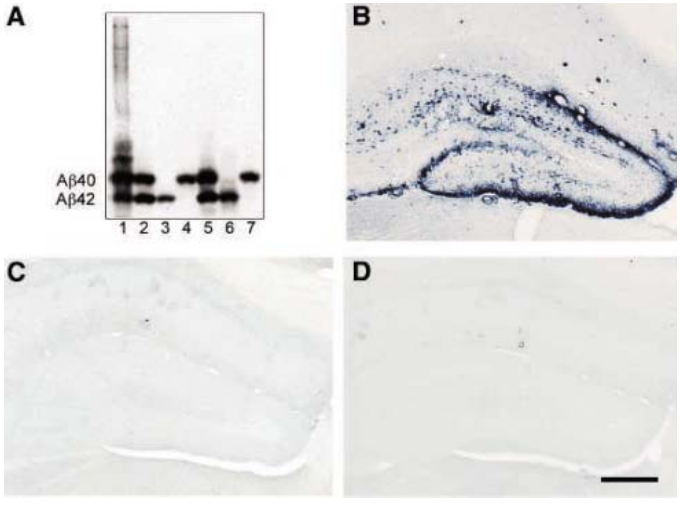

Fig. 4. Fresh and aged Aβ preparations were made from Aβ1-40 or Aβ1-42, or from a 2:1 mixture of Aβ1-40 and Aβ1-42 at ~5 ng/μl, similar to the Aβ concentrations in a 10% APP23 brain extract. (**A**) Urea-based SDS-PAGE followed by immunoblotting with a human Aβ-specific antibody. Lane 1: APP23 Tg brain extract; lane 2: mixed fresh Aβ1-40 + Aβ1-42; lane 3: fresh Aβ1-42; lane 4: fresh Aβ1-40; lane 5: aged Aβ1-40 + Aβ1-42; lane 6: aged Aβ1-42; and lane 7: aged Aβ1-40. Preparations were injected intrahippocampally into 5-month-old male APP23 mice that were analyzed 4 months later. The aged Aβ preparations were fibrillar in nature as verified by electron microscopy and Congo red binding (fig. S6). Multimeric Aβ species are not easily detected with this urea-based gel system, which is designed to separate the different Aβ isoforms. The additional band in the brain extract is likely Aβ1-38. (**B**) Amyloid induction with an APP23 Tg extract. (**C** and **D**) No induced Aβ deposits were detectable with any of the synthetic Aβ preparations at this concentration. Shown are animals injected with freshly mixed Aβ1-40 + Aβ1-42 (C) and aged Aβ1-40 + Aβ1-42 (D). Scale bar, 350 μm.

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Supporting Online Material

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Therapy-Induced Acute Recruitment of Circulating Endothelial Progenitor Cells to Tumors

Yuval Shaked,[1]* Alessia Ciarrocchi,[2]* Marcela Franco,[1] Christina R. Lee,[1] Shan Man,[1] Alison M. Cheung,[3] Daniel J. Hicklin,[4] David Chaplin,[5] F. Stuart Foster,[3] Robert Benezra,[2] Robert S. Kerbel[1]†

The contribution of bone marrow–derived circulating endothelial progenitor cells (CEPs) to tumor angiogenesis has been controversial, primarily because of their low numbers in blood vessels of untreated tumors. We show that treatment of tumor-bearing mice with vascular disrupting agents (VDAs) leads to an acute mobilization of CEPs, which home to the viable tumor rim that characteristically remains after such therapy. Disruption of this CEP spike by antiangiogenic drugs or by genetic manipulation resulted in marked reductions in tumor rim size and blood flow as well as enhanced VDA antitumor activity. These findings also provide a mechanistic rationale for the enhanced efficacy of VDAs when combined with antiangiogenic drugs.

Angiogenesis, the growth of new blood vessels from an existing vasculature, requires expansion of vascular endothelial cells. This can occur locally by proliferation of differentiated endothelial cells, or systemically by mobilization of bone marrow–derived endothelial progenitor cells, which enter the peripheral blood circulation, migrate to sites of angiogenesis, and incorporate into growing vessels (*1*, *2*). The role of such CEPs in tumor angiogenesis is controversial. Estimates of their contribution to the tumor endothelium in untreated tumors range from as much as 10 to 50% (*1*, *2*) to 5% or less (*3*, *4*), with the majority of studies showing figures in the lower range.

To investigate whether the levels of CEP recruitment to the tumor vasculature change during or after certain anticancer therapies, we studied mice treated with VDAs. In contrast to antiangiogenic drugs, which inhibit the formation of new blood vessels, VDAs cause acute occlusion of existing blood vessels, leading to a rapid and massive intratumoral necrosis (*5*, *6*). However, VDA-treated tumors rapidly regrow from a characteristic rim of residual viable cells at the tumor periphery (*5*, *6*) (fig. S1). We hypothesized that a rapid, reactive mobilization and subsequent tumor homing of CEPs might contribute to this regrowth. To test this hypothesis, we first treated non–tumor-bearing BALB/cJ mice with a single dose of a VDA, either combretastin-A4 phosphate (CA4-P) (*5*, *6*) or OXi-4503, a second-generation derivative of CA4-P. Within 4 hours, CEP levels rapidly increased by a factor of 3, returning to basal levels after 24 hours (Fig. 1A and fig. S2A). An OXi-4503 dose range of 25 to 100 mg per kg body weight led to similar increases in viable CEPs (fig. S2B). Total and differential white blood cell (WBC) counts revealed an increase in neutrophils after 4 hours (fig. S2C). Similar changes in CEP levels occurred in other mouse strains (fig. S2D).

Because prior studies have shown that antiangiogenic drugs suppress the mobilization and levels of CEPs (*7*), we reasoned that the VDA-induced spike might be prevented by prior treatment of mice with an antiangiogenic drug such as DC101, a monoclonal antibody to mouse vascular endothelial growth factor receptor–2 (VEGFR-2) (*7*, *8*). Indeed, the CEP spike was not detectable when DC101 was injected into non–tumor-bearing mice 24 hours before OXi-4503 (Fig. 1A). Comparable experiments with MeWo human melanoma–bearing mice yielded similar results (fig. S3A). Three days after treatment with OXi-4503 alone, the tumors in these mice showed a prominent central necrotic area surrounded by a rim of viable tissue 3.3 ± 0.51 mm across. When DC101 was administered 24 hours before OXi-4503, the size of the viable tumor rim was reduced to 1.12 ± 0.33 mm (fig. S3B). Prevention of VDA-induced CEP mobilization may partially explain previous preclinical data showing an increased efficacy of therapies that combine a VDA with an antiangiogenic drug targeting VEGFR-2 (*9*).

We next evaluated the effect of the combination therapy (DC101 plus OXi-4503) on tumor vessel blood perfusion and flow on established (500 mm^3) subdermally implanted MeWo tumors. On day 4, tumors were removed and analyzed for necrosis, hypoxia, perfusion, and functional blood flow [using speckle-variance analysis by high-frequency microultrasound imaging (*10*, *11*)]. As seen in Fig. 1, B and C, and fig. S4, OXi-4503–treated tumors showed a viable and perfused rim encapsulating a central hypoxic and necrotic areas. A single dose of DC101 produced an increase in the hypoxic but not necrotic areas. However, the combination of DC101 and OXi-4503 produced a profound decrease in perfusion along with marked increases in hypoxia and necrosis. There was a surge in functional blood flow in peripheral functional vessels 3 days (but not 4 hours) after OXi-4503 treatment (Fig. 1D and fig. S4D), but this was not observed when DC101 was administered 24 hours before OXi-4503. As expected, the combination of DC101 and OXi-4503 markedly suppressed tumor growth (fig. S5), consistent with a previous study involving different drugs (*9*).

To determine whether the VDA-induced spike in CEPs and WBCs is followed by a preferential homing of the cells to the viable tumor rim, we studied lethally irradiated C57Bl/6J mice transplanted with green fluorescent protein–positive (GFP^+) bone marrow cells. Human tumor cells cannot be grown in such immunocompetent mice, and thus these mice were used as recipients of a subcutaneous injection of syngeneic Lewis Lung carcinoma (LLC) cells (0.5×10^6). The mice were treated with the same drug combination (DC101 and OXi-4053) and schedule when tumors reached 500 mm^3. Untreated and DC101-treated mice showed only a minor incorporation of GFP^+ bone marrow–derived cells into the tumor periphery, consistent with previous reports showing low-level incorporation of bone marrow–derived circulating cells, including CEPs in tumor vessels (*3*, *4*, *7*), whereas OXi-4503 treated mice showed a substantial number of GFP^+ cells at the tumor site, some of which were incorporated into the tumor vasculature, as assessed by staining with CD31 or VEGFR-2 specific antibodies (Fig. 2A and fig. S6, respectively). Prior treatment of the mice with DC101 markedly reduced the number of GFP^+ cells in the tumor periphery (fig. S7). These results suggest that bone marrow–derived cells incorporate into or around the tumor vasculature associated with the viable tumor rim and support the hypothesis that the acute CEP spike observed shortly after OXi-4503 treatment contributes to tumor regrowth. It is possible that other types of proangiogenic bone marrow–derived cells are also recruited to the tumor rim, such as $CD45^+$ (hematopoietic) myeloid/monocytic cell populations (*12*, *13*) (fig. S8).

[1]Department of Molecular and Cellular Biology Research, Sunnybrook Health Sciences Centre, Toronto, Ontario M4N 3M5, Canada. [2]Program of Cancer Biology and Genetics, Memorial Sloan-Kettering Cancer Center, 1275 York Avenue, New York, NY 10021, USA. [3]Department of Imaging Research, Sunnybrook Health Sciences Centre, Toronto, Ontario M4N 3M5, Canada. [4]ImClone Systems, 180 Varick Street, 7th Floor, New York, NY 10014, USA. [5]OXiGENE Inc., 230 Third Avenue, Waltham, MA 02451, USA.

*These authors contributed equally to this work.
†To whom correspondence should be addressed. E-mail: robert.kerbel@sri.utoronto.ca

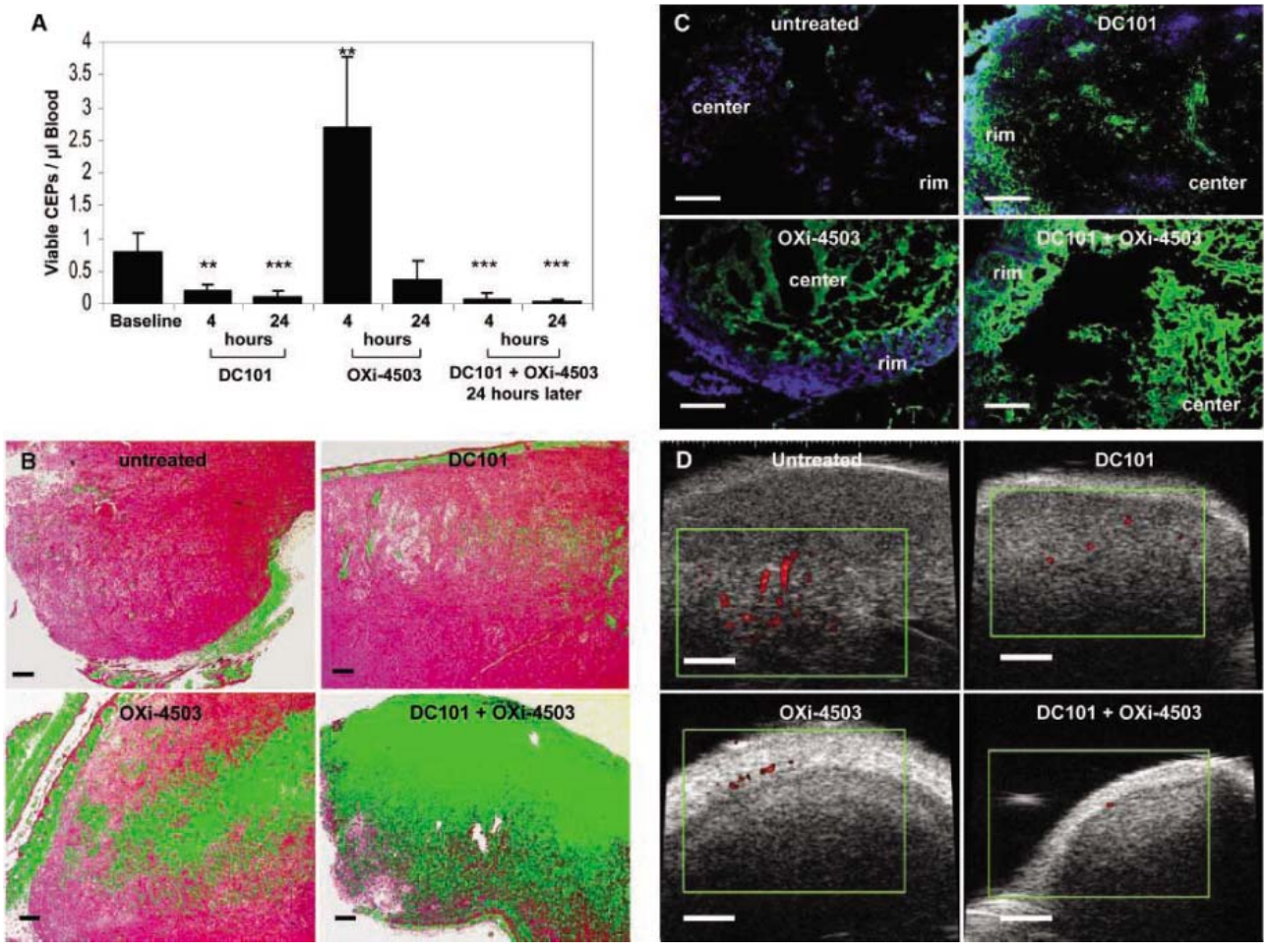

Fig. 1. Elimination of the VDA-induced spike in CEPs, reduction in viable tumor rim blood vessel perfusion as well as blood flow, and increased hypoxia induced by prior treatment with VEGFR-2 monoclonal antibody DC101. (**A**) Eight-week-old non–tumor-bearing BALB/cJ mice ($n = 5$ mice per group) were bled from the retro-orbital sinus 4 and 24 hours after they were treated with OXi-4503 (100 mg/kg), DC101 (800 μg per mouse), or a combination of the two drugs, as indicated. CEP levels were determined using four-color flow cytometry as in (*17*) for each treatment group. Error bars ± SD; $^{**}0.05 > P > 0.01$, $^{***}P < 0.01$. (**B** to **D**) The same drug schedule was used on orthotopically implanted MeWo human melanoma tumor cells. Six- to eight-week-old nude mice were subdermally transplanted with MeWo cells (2×10^6) that were allowed to reach ~500 mm^3, at which point they were treated with DC101 (800 μg), OXi-4503 (100 mg/kg), or DC101 24 hours before 100 mg/kg (at the same dosages). Three days after OXi-4503 treatment, tumors were harvested and evaluated for (B) necrosis (scale bar, 100 μm) and (C) hypoxia (green) and perfusion (blue) (scale bar, 50 μm). In a parallel experiment, mice were evaluated for functional blood flow (D) using high-frequency microultrasound (scale bar, 1 mm). See fig. S4 for summary of quantitative data.

To confirm that the systemic CEP spike contributes significantly to the growth of the viable tumor rim after VDA treatment, we administered OXi-4503 to *Id-1*$^{+/-}$*Id-3*$^{-/-}$ mutant (*Id*^{mut}) mice, which are incapable of mobilizing CEPs (*14*–*16*). As expected, treatment of non–tumor-bearing *Id*^{mut} mice with a single dose of OXi-4503 did not produce the CEP spike that is seen in wild-type mice (Fig. 2B). To evaluate the antitumor activity of VDAs in such mice, we implanted LLC cells subcutaneously into *Id*^{mut} mice and their wild-type controls. In addition, to confirm that the VDA effect is due to a systemic deficiency in CEP mobilization and not primarily to a local angiogenic defect, we "rescued" lethally irradiated *Id*^{mut} mice by transplantation of bone marrow cells derived from wild-type mice as in (*14*); LLC cells were implanted 4 weeks later. When tumor volumes reached 500 mm^3, mice were treated with a single dose of OXi-4503, and 3 days later the tumors were evaluated for levels of necrosis. The tumors in the *Id*^{mut} or wild-type mice transplanted with *Id*^{mut} bone marrow had a reduced viable tumor rim, with most of the tumor consisting of necrotic tissue, whereas the tumors in the wild-type or *Id*^{mut} "rescued" mice showed a more central necrotic area surrounded by a conspicuous thick layer of viable tissue (Fig. 2C and fig. S9). The tumors in *Id*^{mut} mice also showed extensive areas of hypoxia and minimally perfused rims (fig. S10). In the absence of treatment, little tumor necrosis was detected in wild-type or *Id*^{mut} mice, the latter due to the treatment being initiated at early stages of tumor growth. Together, these results reinforce our hypothesis that CEPs mobilized from the bone marrow are a major contributor to the growth of the viable tumor rim after treatment with VDAs.

Our results illustrate that although CEP levels in vessels of untreated tumors are typically low, these levels can suddenly rise in response to acute stress, such as that caused by treatment with a VDA and possibly with other treatments such as maximum-tolerated-dose cytotoxic chemotherapy (*17*). This situation may be analogous to the rapid reactive

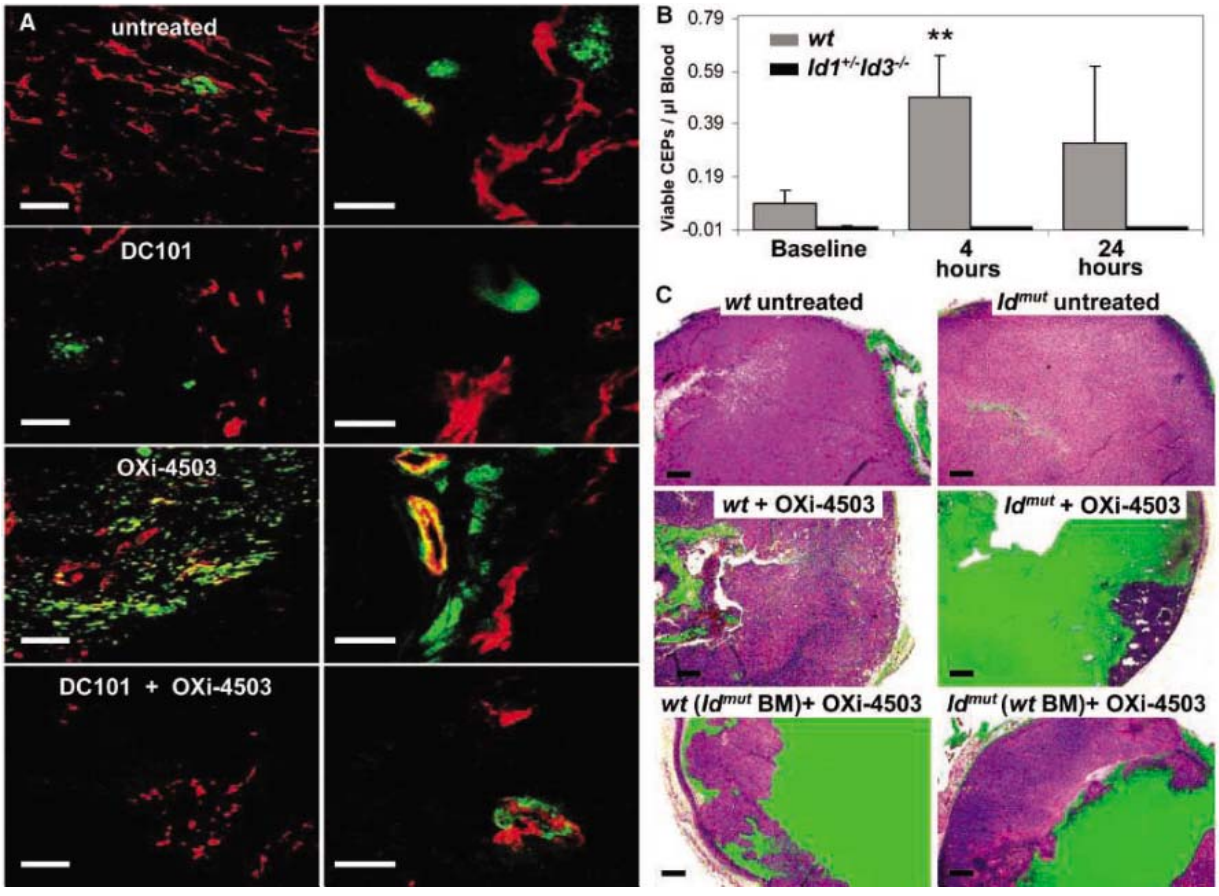

Fig. 2. Homing and incorporation of GFP$^+$ bone marrow–derived cells in mouse tumors after treatment of the tumor-bearing mice with DC101, OXi-4503, or the combination of the two drugs, and effect of OXi-4503 on CEPs and tumors in *Id*^{mut} mice. (**A**) GFP$^+$ bone marrow–transplanted C57Bl/6J mice ($n = 5$ mice per group) were used as recipients of LLC cells (0.5×10^6) injected subcutaneously and treated with DC101, OXi-4503, and the combination of the two drugs at the same doses and schedule described for Fig. 1, B and C; treatment was initiated when tumor volumes reached 500 mm^3. Three days later, tumors were removed and stained for CD31 (red) to mark endothelial cells. GFP$^+$ bone marrow–derived cells are green; scale bars, 50 μm for left images, 20 μm for right images. (**B**) Eight- to 10-week-old non–tumor-bearing *Id*^{mut} mice and their age-matched wild-type (*wt*) controls ($n = 4$ or 5 mice per group) were treated with OXi-4503 (100 mg/kg). Blood drawn from the retro-orbital sinus at baseline, 4 and 24 hours, was processed for viable CEPs as in (*17*). Error bars ± SD; $^{**}0.05 > P > 0.01$. (**C**) Eight- to 10-week-old *Id*^{mut} mice, their age-matched wild-type controls, lethally irradiated *Id*^{mut} mice transplanted with 10^6 bone marrow cells obtained from wild-type control mice, and lethally irradiated wild-type mice transplanted with 10^6 bone marrow cells derived from *Id*^{mut} mice were implanted with LLC cells (0.5×10^7). When tumors reached 500 mm^3, treatment with a single dose of OXi-4503 (100 mg/kg) was initiated. Three days later, mice were killed and tumors were harvested and evaluated for necrosis. Scale bar, 100 μm; BM, bone marrow. See fig. S9 for summary of quantitative data.

mobilization and homing of CEPs to damaged vessels or arteries that occurs after pathological cardiovascular events such as myocardial infarcts (*18*). Our results also provide an additional mechanistic rationale for the enhanced efficacy of VDAs when combined with an antiangiogenic drug (*9*). Finally, they suggest that when a VDA is to be combined with chemotherapy, consideration should be given to the counterintuitive idea of administering chemotherapy shortly after VDA treatment, rather than the opposite sequence, because of the ability of chemotherapy to target CEPs (*17*, *19*).

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Supporting Online Material

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A Genomewide Search for Ribozymes Reveals an HDV-Like Sequence in the Human *CPEB3* Gene

Kourosh Salehi-Ashtiani,[1,2]* Andrej Lupták,[1]* Alexander Litovchick,[1] Jack W. Szostak[1]†

Ribozymes are thought to have played a pivotal role in the early evolution of life, but relatively few have been identified in modern organisms. We performed an in vitro selection aimed at isolating self-cleaving RNAs from the human genome. The selection yielded several ribozymes, one of which is a conserved mammalian sequence that resides in an intron of the *CPEB3* gene, which belongs to a family of genes regulating messenger RNA polyadenylation. The CPEB3 ribozyme is structurally and biochemically related to the human hepatitis delta virus (HDV) ribozymes. The occurrence of this ribozyme exclusively in mammals suggests that it may have evolved as recently as 200 million years ago. We postulate that HDV arose from the human transcriptome.

RNAs play diverse roles in biology, including a variety of regulatory and catalytic functions that together provide support for the "RNA world" hypothesis (*1*). Catalytic RNAs, or ribozymes, include naturally occurring phosphoryl transferases and the ribosomal peptidyl transferase (*2*), as well as in vitro selected RNAs (*3*). Recent discoveries of regulatory cellular ribozymes, such as the bacterial cofactor–dependent ribozyme GlmS (*4*) and the eukaryotic cotranscriptional cleavage (CoTC) ribozyme (*5*), have raised interesting questions about the prevalence and evolution of catalytic RNAs in higher eukaryotes.

Most of the reactions carried out by naturally occurring ribozymes involve the chemistry of the phosphodiester RNA backbone. Among phosphotransferases, self-cleaving ribozymes form perhaps the most diverse subgroup, which includes the hammerhead (*6*), hairpin (*7*), and HDV ribozymes (*8*); the *Neurospora* Varkud satellite (VS) motif (*9*); the bacterial GlmS ribozyme (*4*); the eukaryotic CoTC motif (*5*); and the group I intron–like ribozyme GIR 1 (*10*). Only two of these are associated with mammals: the CoTC motif, the only known self-cleaving human ribozyme, with orthologs in other primates (*5*), and the HDV ribozyme, the only known self-cleaving ribozyme encoded by a human pathogen, with no known evolutionary orthologs. The sparse occurrence of self-cleaving motifs in humans, and mammals in general, could indicate that this class of catalytic RNA has been lost over the course of evolution or, alternatively, that these RNAs have escaped detection. All self-cleaving ribozymes found to date were discovered by careful analysis of transcripts of single genes or RNA genomes of pathogens. Notably, GlmS was first identified as a riboswitch through computational analysis of conserved bacterial RNA structures and was later observed to self-cleave in the presence of a cofactor (*4*). CoTC was identified through detailed studies of cotranscriptional termination of β-globin mRNA synthesis (*5*).

To identify ribozymes encoded in the human genome, we devised an in vitro selection scheme for the isolation of self-cleaving sequences without imposing fixed cleavage target sites. In this scheme, a genomic library is constructed in which library elements are uniform in size [~150 nucleotides (nt)] and are flanked by polymerase chain reaction (PCR) primer sequences [fig. S1 (*11*)]. The library was converted into a single-stranded form, circularized by splint-ligation, and converted back to double-stranded DNA to form a "relaxed" double-stranded circle that can be in vitro transcribed (*11*). Rolling-circle transcription produced tandemly repeated RNAs that preserved the covalent linkage of the entire transcribed sequence, even after self-cleavage (*12*). If a sequence encoded a self-cleaving motif, it could self-cleave under appropriate conditions to produce unit-length copies, as well as kinetically trapped misfolded intermediates such as dimers, trimers, and so on. The size difference between the cleaved multimers and uncleaved sequences provided a positive selection criterion for the enrichment of active molecules. Dimers, which contain one intact copy of the sequence, were isolated by polyacrylamide gel electrophoresis (PAGE) purification, reverse-transcribed, and amplified to reinitiate the cycle.

We started the selection with about 580 ng of RNA (or 10^{15} nucleotides) in concatemeric form. We allowed the RNA to self-cleave for 1 hour in the presence of 5 mM $MgCl_2$ at near-physiological monovalent salt concentration, pH, and temperature (140 mM KCl, 10 mM NaCl, 50 mM Tris-HCl, pH 7.4, 37°C). After separation by denaturing PAGE, RNA fragments corresponding to dimers (~400 nt) were isolated, reverse-transcribed, and amplified to reinitiate the cycle as outlined above. After six rounds of selection, self-cleavage could be detected in the library. Activity increased up to round 12, when the library became dominated by a small number of active sequences that exhibited robust self-cleavage in the presence of physiological concentrations of Mg^{2+} (0.5 to 1 mM $MgCl_2$). The concatemeric RNA of this and preceding active rounds cleaved to form fragments that differed in length by about 200 nt, the unit size of the library. When the kinetically trapped trimers and dimers were isolated and renatured, they self-cleaved to yield smaller products (fig. S1). Thus, the selected library displayed properties in accordance with the experimental design and consistent with the presence of a single cleavage site per monomeric unit.

Cloning and sequencing of the round 12 library (fig. S2) revealed four self-cleaving ribozymes associated with the following human genes: olfactory receptor *OR4K15*, insulin-like growth factor 1 receptor (*IGF1R*), a LINE 1 retroposon, and the cytoplasmic polyadenylation element–binding protein 3 (*CPEB3*) (Fig. 1A and fig. S3). We studied the CPEB3 ribozyme because it occurs within the transcript of a single-copy gene and because it is highly conserved (Fig. 1, B and C).

The isolated CPEB3 clones (including clones from round 10 and 11) originated from 12 independent progenitor sequences (i.e., independent DNase I fragments) of the library (fig. S4). The region of sequence overlap contains the minimal sequence necessary for self-cleavage activity and also delineates the local conservation boundaries among different species in this region of the intron (Fig. 1, C and E). The sequence alignment also revealed several mutations that are not present in the genome. Rather than having randomly accumulated during the repeated PCR amplification cycles, some of these mutations appear to have been highly selected (Fig. 1E and fig. S5). To rule out the possibility that the cleavage activity depends on the mutations that had been accumulated in the course of the selection, we tested the activity of RNA transcribed from a genomic DNA segment amplified directly from human genomic DNA. We tested the putative ribozyme sequence for self-cleavage in the absence of the selection PCR primer sequences and demonstrated that the 209-nt genomic sequence alone was sufficient to carry out self-cleavage (Fig. 1G). Additional 5′ and 3′ flanking sequences do not interfere with the activity of the ribozyme, measured as a first-order rate constant (k_{obs}) of the RNA self-cleavage reaction (k_{obs} = 0.69 ± 0.03 hour^{-1}, $t_{1/2}$ = 1.00 ± 0.04 hours). Furthermore, we designed a set of primers to amplify a 587–base pair (bp) region that included the CPEB3 ribozyme and ~250 bp of flanking sequences on each side. Transcripts made from this genomic segment showed self-cleavage (Fig. 1H). Conversely, shortening the ribozyme to the region of highest conservation allowed us to narrow the sequence to 81 nt, to map precisely

[1]Howard Hughes Medical Institute, Department of Molecular Biology, and Center for Computational and Integrative Biology (CCIB), 7215 Simches Research Center, Massachusetts General Hospital, 185 Cambridge Street, Boston, MA 02114, USA. [2]Center for Cancer Systems Biology (CCSB) and Department of Cancer Biology, Dana-Farber Cancer Institute, 44 Binney Street, Boston, MA 02115, USA.

*These authors contributed equally to this work.
†To whom correspondence should be addressed. E-mail: szostak@molbio.mgh.harvard.edu

the site of cleavage, and to analyze the products of self-cleavage. The 81-nt construct cleaved to yield fragments of 9 nt upstream and 72 nt downstream of the cleavage site. Incubation of the products of the transesterification reaction with polynucleotide kinase (PNK), in the absence of adenosine triphosphate, shifted the mobility of the upstream 9-nt fragment; this change suggested that the upstream fragment contains a 2′-3′ cyclic phosphate and that the downstream 72-nt fragment has a 5′ terminal hydroxyl (Fig. 1I). We confirmed the identity of the products of self-cleavage by noting that the downstream sequence could be phosphorylated at its 5′ end with PNK and that the upstream sequence was not a substrate for T4 RNA ligase. Altogether these experiments demonstrate that the 72-nt core of the sequence is sufficient to carry out self-cleavage, that the ribozyme can function in the presence of its native flanking sequences, and that the chemistry of self-cleavage most likely proceeds via a nucleophilic attack of a 2′ hydroxyl on the adjacent phosphate, which yields a 2′-3′ cyclic phosphate and a 5′ terminal hydroxyl.

BLAST and BLAT analyses (*13*, *14*) demonstrate that the CPEB3 ribozyme sequence, like the *CPEB3* gene itself, is found as a single copy in the genome and is highly conserved in all mammalian species examined, including opossum (a marsupial) (Fig. 1F). The ribozyme resides in a large intron (~46 kbp in human, ~37 kbp in rat, and ~35 kbp in mouse) about

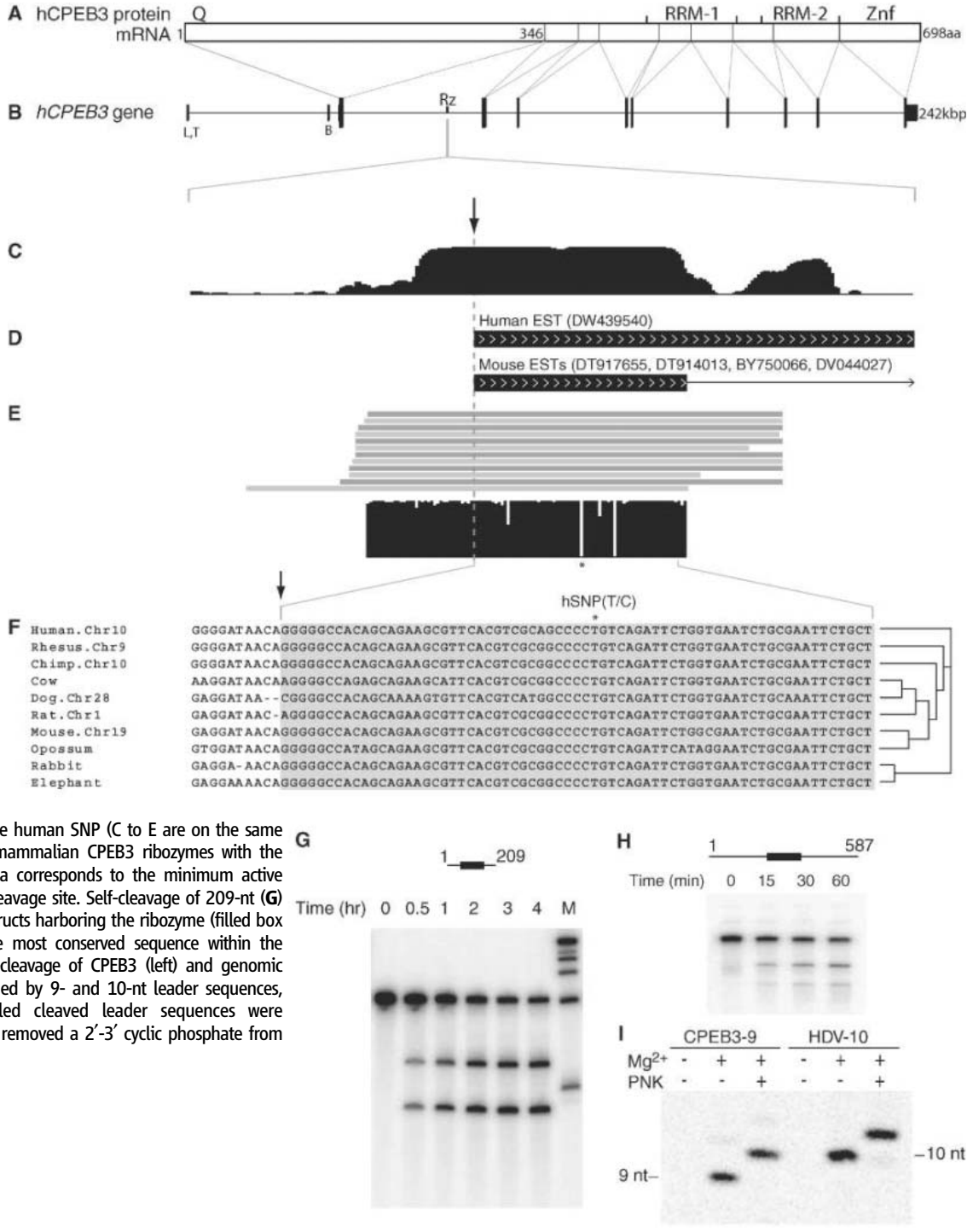

Fig. 1. Mapping and activity of the human CPEB3 ribozyme. (**A**) Primary structure of CPEB3 (also KIAA0940; GenBank NM_014912) protein (Q, glutamine-rich domain; RRM, RNA-binding domains; Znf, zinc finger; and aa, amino acid) and mRNA. Vertical dividers mark splice sites. (**B**) Human *CPEB3* gene. Untranslated tissue-specific exons are marked with letters (L, liver; T, testis; and B, brain tissue) and translated exons with large vertical lines. Rz, location of the self-cleaving sequence in the second intron. (**C**) Mammalian conservation of the self-cleaving sequence. Regions of high conservation show higher amplitude. Arrow indicates the ribozyme cleavage site. (**D**) Human and mouse ESTs that correspond to the region in (C); the 5′ end of the ESTs aligns with the cleavage site of the ribozyme. (**E**) Alignment of the 12 independent isolates of the ribozyme from the in vitro selection (horizontal bars) and conservation of the common sequence. Asterisk marks the position of the human SNP (C to E are on the same scale). (**F**) Alignment of the mammalian CPEB3 ribozymes with the human sequence. Shaded area corresponds to the minimum active sequences; arrow marks the cleavage site. Self-cleavage of 209-nt (**G**) and 587-nt (**H**) genomic constructs harboring the ribozyme (filled box indicates the location of the most conserved sequence within the construct). (**I**) Products of self-cleavage of CPEB3 (left) and genomic HDV (right) ribozymes, preceded by 9- and 10-nt leader sequences, respectively. The 5′-^{32}P-labeled cleaved leader sequences were incubated with T4 PNK, which removed a 2′-3′ cyclic phosphate from the 3′ end of the RNA.

10 to 14 kbp upstream of the second coding exon in all mammals except opossum, where the distance to the exon is about 25 kbp. Because the CPEB3 ribozyme is present in eutheria (placental mammals) and metatheria (marsupials), it is likely to have appeared at least 130 million years ago, before the two groups diverged. We found no ortholog of the ribozyme in orthologs of the *CPEB3* gene in non-mammalian vertebrates, which suggests a mammalian origin for this sequence and indicates that it is likely to have appeared less than ~200 million years ago. The high conservation of the ribozyme sequence among mammals suggests that the sequence is functionally important.

The CPEB3 ribozyme requires divalent metal ions for its cleavage-transesterification. The ribozyme has a relatively low magnesium requirement, half-saturating at ~7.8 mM with a Hill coefficient of 1.4, which suggests that some degree of cooperativity is involved in the metal-dependent catalysis (Fig. 2A). The ribozyme efficiently self-cleaves in the presence of Mn^{2+}, Mg^{2+}, and Ca^{2+}; less efficient cleavage is observed in the presence of Co^{2+}; and no products are detectable in cobalt (III) hexammine, an exchange-inert structural analog of magnesium (II) hexahydrate (Fig. 2B). When subjected to high-resolution PAGE, the products of the reaction comigrate, which suggests a common function for the divalent metal ions in catalysis. Given our data, the simplest model for the metal ion requirement in CPEB3 self-cleavage is that a hydrated divalent metal ion is required to accelerate the transesterification reaction. In some self-cleaving RNAs, such as the hammerhead, hairpin, and VS ribozymes, but not HDV, high concentrations of monovalent metal ions can promote catalysis at rates similar to magnesium-promoted scission (*15*). When we tested the CPEB3 ribozyme in the presence of 3 M Li^+, we did not detect self-cleavage (Fig. 2B), which suggests that CPEB3 ribozyme uses a different catalytic mechanism.

A pH-rate profile of the CPEB3 ribozyme indicates that in 5 mM $MgCl_2$ the activity of the ribozyme is almost constant between pH 5.5 and 8.5, dropping only at higher or lower pHs (Fig. 2C). This profile is consistent with the presence of two functional groups with distinct pK_a values (where K_a is the acid dissociation constant) involved in rate-limiting proton transfers. For comparison, we examined the pH profile of a pool of in vitro selected self-cleaving ribozymes that we had previously isolated from random sequences (*16*). About 83% of this pool consists of new ribozymes, and 17% are diverse hammerhead-like sequences. In contrast to the CPEB3 ribozyme, the activity of this pool increased monotonically with pH, which suggests that acidic apparent pK_a values are not commonly found among self-cleaving ribozymes. Furthermore, when we carried out a cleavage reaction in 80% D_2O, the cleavage rate was less than that in 100% H_2O by a factor of ~1.7. The relatively flat kinetic pH profile and a solvent kinetic isotope effect support a catalytic model in which the rate-limiting step involves at least one proton transfer.

Because the biochemical properties of the CPEB3 ribozyme resemble those of the HDV ribozymes, we next investigated whether these ribozymes have structural similarities. The HDV RNA genome encodes two self-cleaving motifs, which reside in different locations and on opposite strands of the HDV RNA genome and are used to cut nascent rolling-circle replicated RNAs into genome-size copies (*8*, *17*). The ribozymes fold into similar secondary structures characterized by a nested double pseudoknot (*18*, *19*), but, with the exception of several conserved nucleotides, they diverge in primary sequence. The sequence of the CPEB3 ribozyme is not similar to either of the HDV ribozymes; however, the CPEB3 sequence can be folded into an HDV-like secondary structure without violating known HDV structural parameters (Fig. 3). Indeed, the experimentally determined cleavage site of the CPEB3 ribozyme (Fig. 1F) coincides precisely with the cleavage site predicted by this model. Like the HDV ribozyme, the CPEB3 ribozyme can tolerate nucleotide changes immediately upstream of its cleavage site. The homologous sequences in other mammals are all consistent with the HDV-like secondary structure (Fig. 3B). The mutations that appear to have been selectively accumulated during the course of the selection [from 41 sequenced CPEB3 clones (fig. S5)] would result in either conservative changes such as Watson-Crick base pairs to wobble pairs, or changes in a distal loop (Fig. 3B).

To test whether the secondary structure of the CPEB3 ribozyme is HDV-like, we carried out covariation and mutational analyses at CPEB3 positions that are critical for activity in HDV (fig. S7). Disruption of the last base pair of the P3 helix, G19-C27, dramatically lowered the cleavage rate, whereas ribozymes in which the base pair was reversed to C19-G27 retained the wild-type rate. Similarly, in the P1 region, disruption of the third base-pair (G3-C34) to form a C3-C34 mispair lowered the observed cleavage rate by a factor of 200, whereas a G3-G34 pair reduced it by a factor of 4. Reversing the base pair to C3-G34 increased the cleavage rate of the ribozyme by a factor of 2.7 (fig. S7). The 3′ end of the P1 helix forms a wobble pair in the human sequence (C7-A30) and a Watson-Crick pair (C7-G30) in all other mammals (Fig. 3B). Sequences that differ from the human sequence only at this position (e.g., rabbit and elephant, Fig. 1F) appear to self-cleave several times faster, which supports a structural role of the P1 helix.

The 5′ end of the P1 helix is anchored in the ribozyme active site, which retains the 3′ product

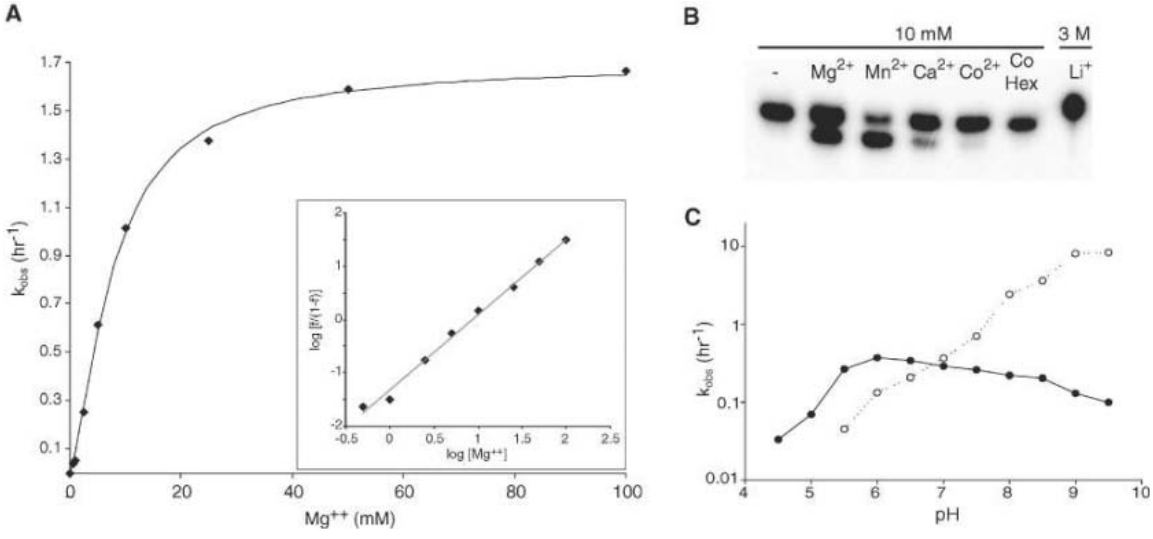

Fig. 2. Biochemical characterization of the CPEB3 ribozyme. (**A**) Mg^{2+} dependence of the self-cleavage kinetics. The cleavage rate reaches half-maximum at 7.8 mM Mg^{2+}. (Insert) Hill analysis of the dependence shows a slope of ~1.4, indicating slight cooperativity in Mg^{2+} binding (*f*; fraction of activity at saturating Mg^{2+}). (**B**) Metal ion requirement for ribozyme activity. Identity and concentration of the metals are indicated above the gel lanes [CoHex, cobalt (III) hexammine]. CPEB3 ribozyme was incubated at 37°C for 10 min. (**C**) pH-rate profile (22°C, 10 mM Mg^{2+}) of the CPEB3 ribozyme (filled circles) and a pool of in vitro selected self-cleaving ribozymes (open circles) (*16*).

of the cleavage reaction (*18*). In the CPEB3 ribozyme, the first nucleotide downstream of the cleavage site forms a wobble pair (G1-U36), as do the corresponding bases in HDV ribozymes. However, the human CPEB3 ribozyme has a single-nucleotide polymorphism (SNP) (U36C), and there are several mammalian orthologs that also form a Watson-Crick base pair at this position and retain cleavage activity (Figs. 1F and 3B and fig. S7).

The secondary structure proposed for CPEB3 ribozyme defines nonhelical elements predicted to be important for RNA self-cleavage. In HDV, a nucleobase, C75 in the genomic and C76 in the antigenomic ribozyme, forms part of the active site and has been proposed to be directly involved in proton shuttling during transesterification (*18*, *20*–*23*). The CPEB3 ribozyme has a nucleobase C57 at the corresponding position. As in the HDV ribozymes, mutation of this residue to uridine or guanosine decreases the cleavage rate by at least a factor of 200 (Fig. 3B).

To define the critical regions of the CPEB3 ribozyme, we performed phosphorothioate interference mapping, in which sulfur substitution for nonbridging oxygens in the RNA backbone can disrupt key interactions within the ribozyme (*24*). The interference mapping of the CPEB3 ribozyme revealed two regions (nucleotides 20 to 23 and 58 to 61 downstream of the cleavage site) where such sulfur substitutions inactivated the ribozyme. Mapping of these positions onto the proposed HDV-like model (fig. S7) placed the phosphorothioate interference sites at positions corresponding to those previously mapped in the genomic HDV ribozyme (*25*) and to solvent-inaccessible backbone positions of the ribozyme core (*26*).

The HDV ribozyme folds into an intricate secondary structure that allows two stacked helical structures, P1-P1.1-P4 and P2-P3, respectively, to pack next to each other (*18*, *22*). The stacked helices are held together on one end by the cross-over strands J1/2 and J3/1. The other end of the P2-P3 helical structure is held in place through the 2 bp of the P1.1 stem, which defines the second nested pseudoknot. The P1.1 minihelix forms one side of the active site of the ribozyme; hence, disruption of this interaction decreases the cleavage rate in both genomic and antigenomic HDV ribozymes (*19*). It is surprising that the CPEB3 ribozyme only forms 1 bp of the P1.1 interaction (C22-G37). The second pair forms a weak U21-U38 interaction that can be strengthened with a U38A mutation, thus forming a 2-bp P1.1 and increasing the cleavage rate by almost 10-fold (Fig. 3B).

There are five expressed sequence tags (ESTs) (one from human liver and four from three different murine tissues) in public databases that overlap with the conserved region of the self-cleaving sequence, all of which share the same 5′ terminal sequence that maps to the CPEB3 ribozyme cleavage site (Fig. 1D). The existence of these ESTs suggests that the CPEB3 ribozyme is active in vivo and that the downstream product of self-cleavage is sufficiently stable that it can be subsequently detected by EST mapping. To investigate in vivo expression and self-cleavage of the ribozyme, we used reverse transcriptase (RT)–PCR to analyze human and murine total RNA extracts prepared in the absence of divalent metal ions from several tissues (fig. S9) (*11*). Mapping by rapid amplification of 5′ cDNA ends (5′ RACE) of those extracts revealed that the site of self-cleavage is the same as in the ESTs and at the position mapped in vitro, which together indicate that the ribozyme is expressed and self-cleaves in vivo (fig. S8).

The CPEB3 ribozyme is an example of a class of self-cleaving ribozyme previously seen only in the HDV. The HDV ribozymes are structurally (*18*, *22*) and informationally (*27*, *28*) complex. Thus, in contrast to the hammerhead fold (*16*), the HDV fold is unlikely to have arisen independently multiple times. In further support of this suggestion, we found no sequences that could fold into an HDV-like secondary structure among the self-cleaving sequences isolated from a random pool (*16*) or among conserved mammalian sequences. On the basis of these observations, we hypothesize that HDV ribozymes and the CPEB3 ribozyme are evolutionarily related and that HDV, a single-stranded RNA satellite virus for hepatitis B (HBV) infection, arose from the human transcriptome. The reverse scenario—i.e., that the CPEB3 ribozyme came from HDV—is less likely. In order for HDV to transfer the ribozyme to mammals, the transfer would have to have occurred early in mammalian evolution (~200 million years ago), and ancestral versions of HDV and HBV would have to have existed as well. However, although HBV has been isolated from other animals, even nonmammals, and may have ancient origins, HDV has been isolated only from human tissues. The absence of HDV isolates from other animals suggests that HDV is of recent origin and that it arose from the human transcriptome, acquiring both the delta antigen protein (*29*) and the self-cleaving ribozyme from its host. Furthermore, if the HDV fold is exclusive to mammalian genomes and pathogens, then it is unlikely to be a descendant of the RNA world, which implies that structurally complex ribozymes have evolved in modern protein-dominated organisms. The rapidly evolving intron sequences may have been a fertile ground for the emergence of novel regulatory RNAs.

The CPEB3 ribozyme seems to be optimized to cleave more slowly than the HDV ribozymes; this inference is based on the conservation of a weak P1.1 stem among all mammalian sequences. We speculate that this slow cleavage activity allows normal splicing of the CPEB3 pre-mRNA to occur under basal conditions. The ribozyme is located about 11.5 kbp upstream of the third CPEB3 exon, so that a transcribing RNA polymerase will take about 10 min to reach the next intron-exon junction after synthesizing the CPEB3 ribozyme (*30*). Thus, most of the time, the spliceosome will join the exons before intron

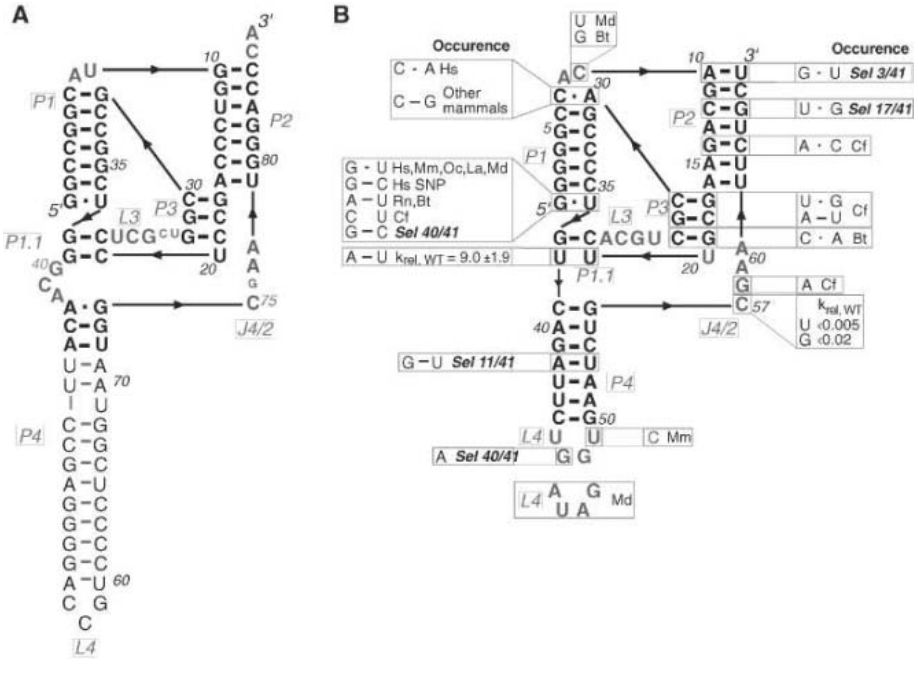

Fig. 3. Secondary structure of the genomic HDV ribozyme (**A**) and the human CPEB3 ribozyme (**B**). Names of ribozyme segments are boxed (P, paired; L, loop; and J, joining region). Numbers represent positions in the chain relative to the cleavage site (marked by 5′). (B) Variants of the CPEB3 ribozyme found in mammals (Hs, human; Mm, mouse; Rn, rat; Oc, rabbit; Cf, dog; La, elephant; Bt, cow; and Md, opossum) and among 41 sequenced clones from the human genomic selection described in the text (Sel). In vitro self-cleavage rates of U38A and C57 mutants are shown relative to wild-type ribozyme.

cleavage. If, however, the rate of ribozyme self-cleavage is up-regulated, the number of truncated CPEB3 pre-mRNAs could increase significantly. The CPEB3 ribozyme sequences in all species examined retain the weak P1.1 interaction, which suggests that slow cleavage is a positively selected feature of this ribozyme. Because there are many possible ways to decrease the cleavage rate, conserving this specific way of lowering the rate suggests that a trans-acting factor interacting at or near this site could readily stimulate the activity of the ribozyme. Rapid degradation of the cleaved RNA fragments could switch off the expression of the CPEB3 protein. Alternatively, cleavage of the CPEB3 pre-mRNA, perhaps followed by subsequent processing events, might produce smaller mRNAs that could be translated into truncated versions of the protein.

CPEB3 is a member of a protein family that regulates local polyadenylation of mRNAs in the cytoplasm of, among other tissues, neurons and oocytes (*31*). The CPEB3 full mRNA has been detected in human brain, skeletal muscle, and heart and to a lesser degree in liver, kidney, testis, and ovary tissues (*32*, *33*). In mouse hippocampus, the gene is up-regulated transiently after induction of seizure, and it has been implicated in long-term potentiation (*33*). CPEB3 differs from other members of the CPEB family by its glutamine-rich N-terminal domain, which is encoded upstream of the CPEB3 ribozyme (Fig. 1, A and B). The function of this domain is unknown, but the glutamine-rich N-terminal domain of the *Aplysia* CPEB protein has been shown to aggregate into a prionlike structure that in turn makes the protein more active in promoting polyadenylation and that may play a role in the molecular basis for long-term facilitation (*34*, *35*).

The four ribozymes isolated from our selection most likely represent only a small fraction of all cellular ribozymes. Our selection operated under a number of constraints including defined limits for size, cleavage rate, and selectable biochemical properties. For instance, ribozymes larger than ~150 nt could not have been isolated from our library. By changing these parameters and by including cellular factors such as small molecules, it may be possible to isolate additional genomic ribozymes, some of which may be modulated by cofactors, as is the case with the recently discovered bacterial cofactor–dependent ribozyme and the eukaryotic CoTC ribozyme (*4*, *5*).

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Supporting Online Material

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Epilepsy-Related Ligand/Receptor Complex LGI1 and ADAM22 Regulate Synaptic Transmission

Yuko Fukata,[1] Hillel Adesnik,[2] Tsuyoshi Iwanaga,[1] David S. Bredt,[3] Roger A. Nicoll,[2] Masaki Fukata[1,4]*

Abnormally synchronized synaptic transmission in the brain causes epilepsy. Most inherited forms of epilepsy result from mutations in ion channels. However, one form of epilepsy, autosomal dominant partial epilepsy with auditory features (ADPEAF), is characterized by mutations in a secreted neuronal protein, LGI1. We show that ADAM22, a transmembrane protein that when mutated itself causes seizure, serves as a receptor for LGI1. LGI1 enhances AMPA receptor-mediated synaptic transmission in hippocampal slices. The mutated form of LGI1 fails to bind to ADAM22. ADAM22 is anchored to the postsynaptic density by cytoskeletal scaffolds containing stargazin. These studies in rat brain indicate possible avenues for understanding human epilepsy.

Physiological functioning of the mammalian brain involves a finely tuned balance between excitation and inhibition in neural circuits. Upsetting this delicate balance can cause epilepsy, which is a devastating and poorly treated disease. Because many genes that cause epilepsies encode synaptic ion channels, characterization of synaptic protein complexes in rat brain can provide essential insights into molecular mechanisms underlying epilepsy.

The postsynaptic density-95 (PSD-95) is a scaffolding protein at excitatory synapses and plays critical roles in synaptogenesis and synaptic plasticity (*1*–*5*). PSD-95 contains an array of protein/protein interaction domains and forms protein complexes with various synaptic proteins, which help organize AMPA (α-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid) and NMDA (*N*-methyl-D-aspartate)–type glutamate receptors and cell adhesion molecules at synapses. Immunoprecipitation of PSD-95 from rat brain extracts resulted in selective purification of proteins with molecular masses of 95 kD (p95) and 60 kD (p60) (Fig. 1A). Mass spectrometry indicated that p95 contained PSD-95 and ADAM22 (*6*), and that p60 was LGI1 (*7*–*10*) (table S1). Western blotting showed that stargazin (*11*–*13*), a transmembrane AMPA receptor (AMPAR) regulatory protein, also coprecipitated (Fig. 1B). The recovery of ADAM22,

[1]Laboratory of Genomics and Proteomics, National Institute for Longevity Sciences, National Center for Geriatrics and Gerontology, Obu, Aichi 474-8522, Japan. [2]Department of Cellular and Molecular Pharmacology, University of California at San Francisco, San Francisco, CA 94143, USA. [3]Department of Integrative Biology, Eli Lilly and Company, Indianapolis, IN 46285, USA. [4]PRESTO, Japan Science and Technology Agency, Kawaguchi, Saitama 332-0012, Japan.

*To whom correspondence should be addressed. E-mail: fmasaki@nils.go.jp

LGI1, and stargazin showed similar efficiency (Fig. 1B, ~10% of input). In contrast, other reported PSD-95–binding proteins, neuroligin and NR1, were hardly detected under our conditions. The interaction of ADAM22 and LGI1 with PSD-95 is specific, as PSD-95 and LGI1 quantitatively coimmunoprecipitated with ADAM22 (fig. S1 and table S1). Synapse-associated protein 102 (SAP102), another postsynaptic scaffolding protein, did not interact with ADAM22 and LGI1.

All three proteins that associate with PSD-95 in our immunoprecipitation are genetically linked to epilepsy. Stargazin is mutated in stargazer mice with absence epilepsy and ataxia (*14*), and stargazin regulates AMPAR trafficking and gating as an auxiliary subunit (*11*–*13*). LGI1 is a secreted neuronal protein (*7*), and its mutations have been found in patients with autosomal dominant partial epilepsy with auditory features (ADPEAF) (*8*, *9*, *15*). ADPEAF is a rare form of familial idiopathic lateral temporal lobe epilepsy characterized by partial seizures with auditory disturbances (OMIM 600512). ADAM22 shares homology to a large family of transmembrane ADAM metalloproteases but is catalytically inactive (*6*, *16*) and is considered either a cell adhesion molecule or an orphan receptor (*16*). ADAM22-deficient mice show cerebellar ataxia and die around 2 to 3 weeks after birth because of multiple seizures (*6*).

To understand the function of this PSD-95–associated protein complex, we defined the modes for interaction. The C-terminal tail of stargazin binds to the first two PDZ domains of PSD-95 (*17*) (figs. S3A and S3B), and we found that one of the ADAM22 splicing variants has a C-terminal PDZ binding motif (-Glu-Thr-Ser-Ile, -ETSI) that interacts selectively with the C-terminal half containing the third PDZ domain of PSD-95 (Fig. 2A and fig. S3B). LGI1 has an N-terminal signal sequence (Fig. 2A) and is secreted from transfected hippocampal neurons (fig. S6C) and from transfected HEK293 cells (*7*, *18*) (fig. S4, A and B) as an oligomer (fig. S4C). As PSD-95 occurs on the inner surfaces of postsynaptic membranes, extracellular LGI1 must interact with a transmembrane protein in the PSD-95 immunoprecipitates. Using cDNA transfection, we found that ADAM22, but not stargazin, specifically interacted with LGI1 (fig. S4D). Furthermore, we found that transfected LGI1, ADAM22, and PSD-95 form a tripartite complex (Fig. 2B). As further evidence for this interaction, we stained cells without permeabilization and found that LGI1 interacts specifically with ADAM22 on the cell surface, indicating that secreted LGI1 binds to the ectodomain of ADAM22. As expected, our extracellular domain binding assay readily detects the interaction of Slit2 with its receptor Robo2 (*19*) (Fig. 2C).

LGI1 has two structural domains, LRR (leucine-rich repeat) and EPTP (Epitempin) repeat (*20*, *21*) (Fig. 2A). The LRR domains show high homology to Slit, a repulsive ligand

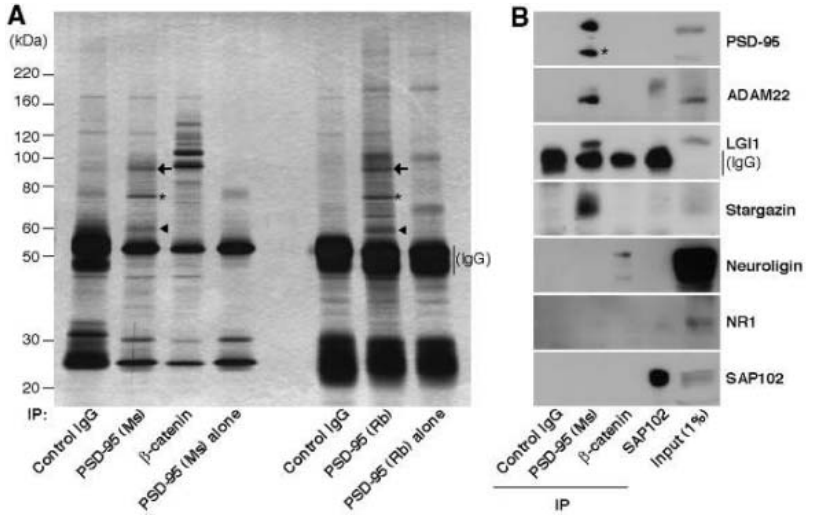

Fig. 1. Identification of a PSD-95–associated protein complex containing ADAM22 and LGI1. (**A**) Immunoprecipitation of PSD-95 from adult rat brain extracts showed a series of bands. Specific bands shared by two independent PSD-95 antibodies were identified by mass spectrometry. p95 (by arrows) contained PSD-95 and ADAM22, and p60 (by arrowheads) was LGI1. A PSD-95 degradation product (p75) is shown with asterisks. IP, immunoprecipitation. (**B**) Western blotting showed that ADAM22, LGI1, and stargazin specifically coprecipitated with PSD-95.

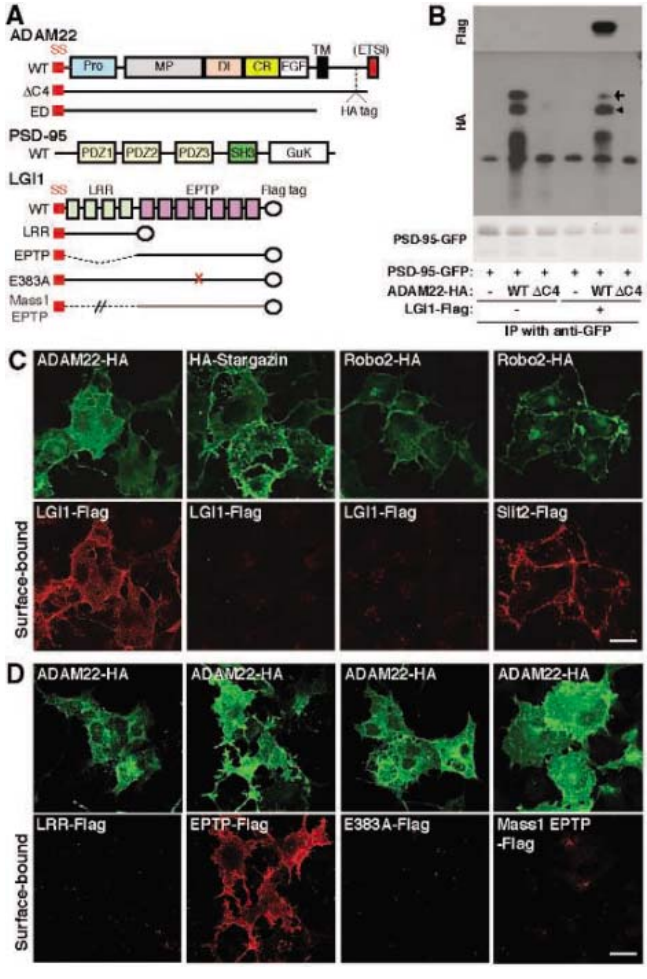

Fig. 2. Tripartite complex formation of PSD-95, ADAM22, and LGI1. (**A**) Domain structures of ADAM22, PSD-95, and LGI1. SS, signal sequence; Pro, prodomain; MP, inactive metalloprotease domain; DI, disintegrin domain; CR, cysteine-rich domain; EGF, EGF-like domain; TM, transmembrane domain. ETSI represents the type I PDZ binding motif of ADAM22. WT, wild type; ΔC4, missing ETSI; ED, extracellular domain; GuK, guanylate kinase domain. LRR, leucine-rich repeat; EPTP, Epitempin repeat; E383A, a point mutant changing Glu (amino acid 383 in the fourth EPTP repeat) to Ala. (**B**) Tripartite complex of PSD-95/ADAM22/LGI1. PSD-95-GFP and ADAM22-HA were cotransfected with or without LGI1-Flag, and PSD-95-GFP was immunoprecipitated (lowest panel, stained by Coomassie brilliant blue). LGI1 indirectly binds to PSD-95 through ADAM22. An arrow and an arrowhead indicate the position of immature and mature ADAM22, respectively. (**C** and **D**) Interaction between secreted LGI1 and ADAM22 on the cell surface. Indicated cDNAs were cotransfected into COS7 cells. At 24 hours after transfection, surface-bound Flag-tagged proteins (red) were labeled before cell permeabilization, and then HA-tagged proteins were stained (green). The EPTP domain of LGI1 mediates ADAM22 binding. LGI1 E383A, an ADPEAF mutant, failed to bind to ADAM22. Scale bars, 10 μm.

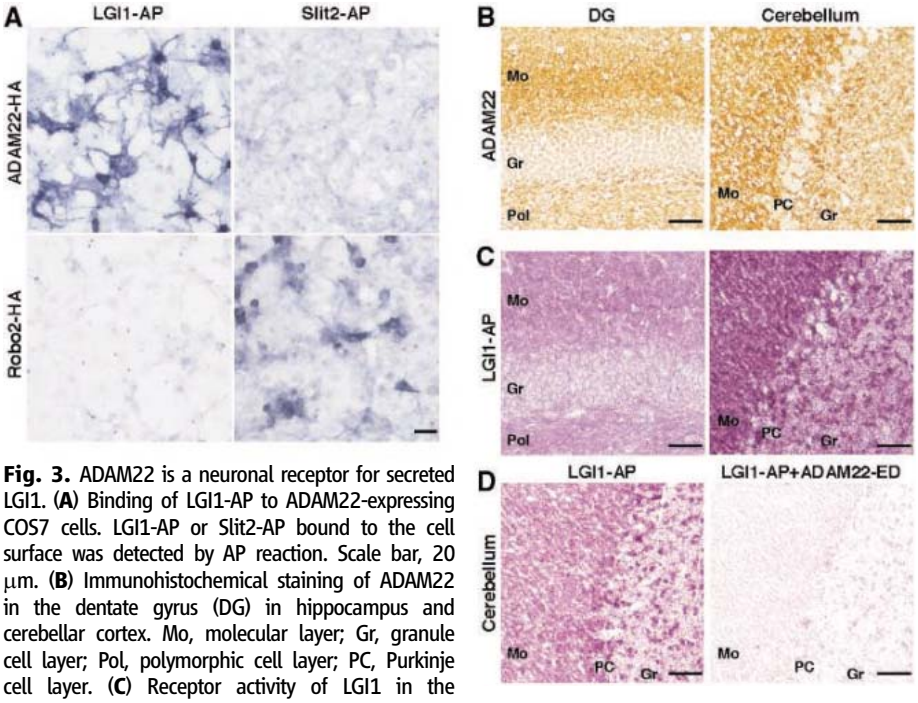

Fig. 3. ADAM22 is a neuronal receptor for secreted LGI1. (**A**) Binding of LGI1-AP to ADAM22-expressing COS7 cells. LGI1-AP or Slit2-AP bound to the cell surface was detected by AP reaction. Scale bar, 20 μm. (**B**) Immunohistochemical staining of ADAM22 in the dentate gyrus (DG) in hippocampus and cerebellar cortex. Mo, molecular layer; Gr, granule cell layer; Pol, polymorphic cell layer; PC, Purkinje cell layer. (**C**) Receptor activity of LGI1 in the hippocampus and cerebellar cortex. Mouse brain sections were treated with conditioned media containing LGI1-AP. LGI1 receptor activity corresponded to the regions where ADAM22 is expressed. (**D**) Preincubation of LGI1-AP with the soluble form of ADAM22 (ADAM22-ED) significantly blocked the LGI1-AP binding in cerebellar cortex. Scale bars in (B), (C), and (D), 50 μm.

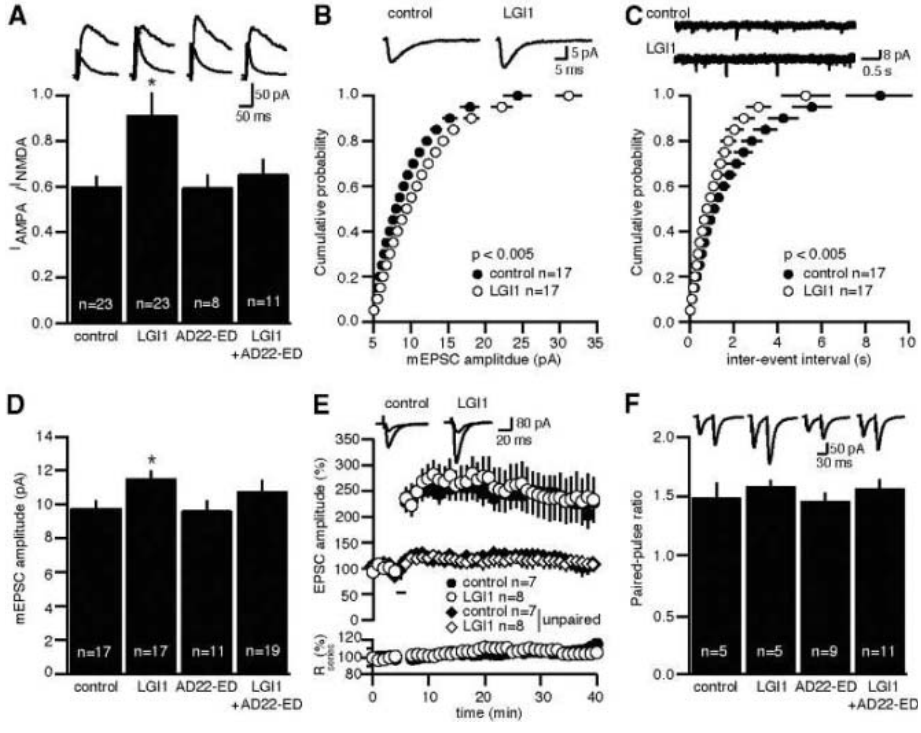

Fig. 4. LGI1 selectively enhances AMPAR-mediated synaptic currents. (**A**) Incubation of slices in buffer containing LGI1 media significantly increased the synaptic AMPA/NMDA ratio ($P < 0.05$), and the effect was blocked by preincubation of LGI1 with the soluble form of ADAM22 (AD22-ED). (**B** to **D**) Incubation of hippocampal slices with LGI1-containing media increases synaptic AMPA receptor numbers. (B) Cumulative distribution plot of mEPSCs from cells in slices incubated in LGI1 as compared with control ($P < 0.005$). (C) Cumulative distribution plot of the interevent interval of mEPSCs in the same cells as in (B) ($P < 0.005$). (D) The increase in mEPSC amplitude by LGI1 was reduced by preincubation with the extracellular domain of ADAM22. (**E**) LGI1 incubation does not alter the magnitude of pairing-induced whole-cell LTP. (**F**) No change in paired-pulse ratio is seen in slices incubated in LGI1, ADAM22-ED, or both ($P = 0.68$).

for the Robo receptor (*19*); the EPTP repeat domain is shared with Mass1/VLGR/USH2C, genes that cause audiogenic epilepsy in Fringe mice and Usher syndrome in humans (*15*, *20*–*23*) (OMIM 602851). We found that the EPTP domain (amino acids 224 to 557) mediates LGI1 binding to ADAM22. The point mutation (E383A) observed in ADPEAF (*8*) prevented its secretion (*7*) (figs. S4, B and C) and binding to ADAM22 (Fig. 2D and fig. S4E). ADAM22 did not interact with another EPTP domain (amino acids 3194 to 3530) of Mass1/VLGR/USH2C. We also found that LGI1 bound to ADAM23, the closest homolog of ADAM22, but not to the more distantly related ADAM9 (fig. S5). The disintegrin domain of ADAM22 is essential for LGI1 binding, as ADAM22(D509N) harboring a mutation in its disintegrin domain did not bind to LGI1 (fig. S5A).

To demonstrate directly the receptor/ligand relationship of ADAM22/LGI1, we constructed a secreted alkaline phosphatase (AP) fusion protein of LGI1 (LGI1-AP). LGI1-AP bound to the surface of cells only when transfected with ADAM22 (Fig. 3A). Slit2-AP did not bind to ADAM22-transfected cells. Under the conditions, Slit2-AP specifically bound to Robo2-transfected cells (*19*). To test whether the interaction of LGI1 with ADAM22 is stoichiometric, the ADAM22 immunoprecipitate from brain was evaluated by Coomassie blue staining and quantitative Western blotting. The stoichiometry of LGI1 binding to ADAM22 was at least 1.0 (fig. S6, A and B). Furthermore, the secreted LGI1 accumulated with ADAM22 at synaptic puncta in hippocampal neurons, where PSD-95 was localized (fig. S6, C and D). Taken together, these results imply that secreted LGI1 serves as a specific extracellular ligand for ADAM22 and that the LGI1/ADAM22 complex is scaffolded by PSD-95.

LGI1 mRNA is coexpressed with ADAM22 and PSD-95 mRNAs in hippocampus, cerebellum, and cerebral cortex (fig. S7, A to C). Immunohistochemical analysis with a specific antibody (fig. S7D) showed that ADAM22 protein occurs in hippocampus and cerebellum (Fig. 3B). We used the LGI1-AP fusion to detect LGI1 receptor activity directly in brain. LGI1-AP detected high receptor activity in the hippocampus and cerebellar cortex (Fig. 3C and fig. S7E). The molecular layers of dentate gyrus (DG) and CA1 regions in the hippocampus were labeled. In the cerebellar cortex, labeling occurred in neuropil of the molecular layer and synaptic glomeruli of the granular layer. These regions corresponded to the regions where ADAM22 is expressed. Preincubation of LGI1-AP with the soluble extracellular domain of ADAM22 (ADAM22-ED, depicted in Fig. 2A) inhibited LGI1-AP binding (Fig. 3D and fig. S7E), consistent with ADAM22 being a receptor for LGI1.

Because PSD-95 controls synaptic AMPA receptor number (*4*), we next asked whether application of LGI1 to hippocampal slices would influence glutamatergic transmission. Incubation

of hippocampal slices in LGI1-AP significantly increased the synaptic AMPA/NMDA ratio (Fig. 4A) (control = 0.60 ± 0.046; LGI1 = 0.90 ± 0.10; n = 23 for each group; $P < 0.05$); nontagged LGI1 showed a similar effect, so we pooled the data. The effects of LGI1 on synaptic currents could be prevented by preincubation of LGI1 with ADAM22-ED (Fig. 4A) (ADAM22-ED = 0.59 ± 0.06; LGI1+ADAM22-ED = 0.65 ± 0.07), suggesting that an interaction between LGI1 and ADAM22 was required for the increased AMPA/NMDA ratio. To determine whether LGI1 directly affects the number of synaptic AMPARs, we measured AMPAR-mediated spontaneous miniature excitatory postsynaptic currents (mEPSCs). LGI1 incubation increased the average amplitude of these events (n = 17, both groups, $P < 0.005$), and preincubation of LGI1 with ADAM22-ED blocked this increase (Fig. 4, B and D). The frequency of spontaneous events was also increased, likely due to the increased detection of enlarged events that now reached threshold (Fig. 4C) (n = 17, each group, $P < 0.005$). Supporting the electrophysiological recording, LGI1 expression significantly increased AMPA receptor surface expression in cultured hippocampal neurons (fig. S8).

Might the potentiation of synaptic AMPA currents by LGI1 share a mechanism with long-term potentiation (LTP), an activity-dependent process that involves synaptic insertion of AMPARs? To address this, we determined whether LGI1 incubation occludes LTP. No significant change in LTP induction was found between control and LGI1-treated slices (Fig. 4E) (control = 247 ± 30%, n = 7; LGI1 = 242 ± 43%, n = 8 at 30 min after LTP induction, $P = 0.93$), suggesting that LGI1 strengthens excitatory synapses by a mechanism distinct from LTP. Finally, we tested whether LGI1 incubation affects presynaptic properties by measuring paired-pulse facilitation. No difference was found between control and LGI1-treated groups (Fig. 4F) (control = 1.50 ± 0.46, LGI1 = 1.58 ± 0.05, ADAM22-ED = 1.45 ± 0.07, LGI1+ADAM22-ED = 1.56 ± 0.08, $P = 0.68$). Taken together, our data indicate that the effects of LGI1 on synaptic transmission are exclusively postsynaptic.

This study establishes a neuronal ligand-receptor interaction between LGI1 and ADAM22, both of which are genetically related to epilepsy. This study also identifies LGI1 as an extracellular factor that controls synaptic strength at excitatory synapses. Stargazin controls the trafficking and gating of AMPARs, and PSD-95 anchors the AMPAR/stargazin complex at postsynaptic sites (*11*). Because the ADAM22 and stargazin binding sites on PSD-95 do not overlap, the LGI1/ADAM22 complex may stabilize the AMPAR/stargazin complex on the PSD-95–scaffolding platform (fig. S9). Supporting the idea, ADAM22 interacted with stargazin through PSD-95 (fig. S3C). Very recently, LGI1 was reported to be a subunit of Kv1.1-containing voltage-gated potassium channels and to inhibit channel inactivation by a cytoplasmic regulatory protein, Kvβ1 (*24*). As LGI1 is secreted, it remains unclear how it might modulate a cytosolic potassium channel mechanism.

This study defines a potentially general mode for protein-protein interaction between EPTP domains and the ectodomain of some ADAM family proteins. LGI4, another member of the LGI family, is mutated in *claw paw (clp)* mice, which show hypomyelination throughout the peripheral nervous system (*18*). Mice lacking ADAM22 display similar hypomyelination of the peripheral nervous system (*6*). Knockouts of Mass1/VLGR/USH2C, ADAM23 (*25*), or ADAM22 (*6*) all display a seizure phenotype. These phenotypes are consistent with EPTP domain interactions with ectodomains of certain ADAM family proteins. Future binding and structural analysis will be needed to clarify the nature of the EPTP domain/ADAM family interaction. This epileptic ligand/receptor complex LGI1/ADAM22 could become a therapeutic target for synaptic disorders.

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PirB Restricts Ocular-Dominance Plasticity in Visual Cortex

Josh Syken, Tadzia GrandPre, Patrick O. Kanold, Carla J. Shatz*

Experience can alter synaptic connectivity throughout life, but the degree of plasticity present at each age is regulated by mechanisms that remain largely unknown. Here, we demonstrate that Paired-immunoglobulin–like receptor B (PirB), a major histocompatibility complex class I (MHCI) receptor, is expressed in subsets of neurons throughout the brain. Neuronal PirB protein is associated with synapses and forms complexes with the phosphatases Shp-1 and Shp-2. Soluble PirB fusion protein binds to cortical neurons in an MHCI-dependent manner. In mutant mice lacking functional PirB, cortical ocular-dominance plasticity is more robust at all ages. Thus, an MHCI receptor is expressed in central nervous system neurons and functions to limit the extent of experience-dependent plasticity in the visual cortex throughout life. PirB is also expressed in many other regions of the central nervous system, suggesting that it may function broadly to stabilize neural circuits.

Plasticity of connections during development is thought to be driven by cellular processes that strengthen or weaken existing synapses in response to neuronal activity, followed by long-term structural alterations to circuits. The cellular and molecular machinery responsible for synaptic plasticity are well studied (*1*, *2*), but the mechanisms and molecules that couple short-term synaptic changes to long-term structural remodeling are less understood.

One family of proteins that is important for activity-dependent structural remodeling of neural circuits during development is MHCI (*3*).

Department of Neurobiology, Harvard Medical School, 220 Longwood Avenue, Boston, MA 02115, USA.

*To whom correspondence should be addressed. E-mail: carla_shatz@hms.harvard.edu

This family of transmembrane cell surface proteins was thought to act exclusively in cellular recognition by the immune system. However, MHCI genes are now known to be expressed in neurons, where they are regulated by neuronal activity (*3*, *4*), and by adenosine 3′,5′-monophosphate response element–binding protein (CREB) (*5*) and are essential for normal synaptic plasticity (*3*). MHCI proteins function through interactions with a variety of transmembrane receptors on immune system cells (*6*, *7*). These interactions are the means by which normal cells are distinguished from abnormal or foreign cells. In the nervous system, the mechanisms by which neuronal MHCI modulates synaptic development are not understood. One hypothesis inspired by examples from the immune system is that neuronal MHCI functions by engaging transmembrane MHCI receptors expressed on other neurons. These interactions could generate intracellular signals that ultimately alter synaptic strength, neuronal morphology, and circuit properties. One such MHCI receptor, known from its role in regulating immune cell activation, is PirB (*8*–*13*).

To examine whether PirB is expressed in the brain, we performed in situ hybridization using PirB-specific probes on mouse brain sections of various ages. Specific mRNA signal was detected throughout the brain at all ages tested, with strong expression in the cerebral cortex, hippocampus, cerebellum, and olfactory bulb (Fig. 1, A to C). These brain regions are also known to express MHCI mRNA and protein (*3*, *4*). PirB protein is also expressed in the brain, as revealed by immunostaining sections with antibodies specific for the cytoplasmic domain of PirB (Fig. 1, D and E). Protein is detected on subsets of neuronal cell bodies (such as those located in cortical layers 5 and 6), on hippocampal and cerebellar neurons, on axonal pathways, and within neuropil.

To confirm that PirB protein is expressed in neurons, neocortex from embryonic day 15 (E15) or postnatal day 0 (P0) mice was dissociated, grown in vitro, and immunostained with one of three different PirB-specific antibodies, in conjunction with neuron-specific markers (Fig. 1, F to J). In these cultures, about 20 to 50% of the neurons are immunoreactive for PirB. Immunostaining is present in axonal growth cones, localized behind the actin-rich leading edge and the zone of synapsin-immunostained vesicles (Fig. 1F). PirB protein is enriched in neuronal processes (Fig. 1, G to J), where it appears as puncta. PirB immunostaining is often very close to, but rarely overlaps with, the presynaptic proteins synaptophysin or synapsin (Fig. 1, H to J), suggesting that PirB is localized at or near synapses. Together, these experiments demonstrate that PirB is expressed by central nervous system neurons both in vivo and in vitro.

Because PirB binds to MHCI in immune cells (*10*, *11*), we investigated whether PirB protein can bind to MHCI on neurons. For these experiments, we generated a soluble recombinant fusion protein consisting of the extracellular domain of PirB fused to alkaline phosphatase (PirB-AP). First, we verified that this reagent binds to cells in an MHCI-dependent manner using cultured mouse embryo fibroblasts (MEFs) derived from wild-type mice or from mice with deleted β2-microglobulin and Tap1 genes (β2m/Tap$^{-/-}$), in which surface expression of MHCI proteins is reduced (*14*). PirB-AP binds to wild-type MEFs, and this binding is markedly reduced on β2m/Tap$^{-/-}$ cells (Fig. 2A), indicating that PirB-AP does indeed bind to MHCI. Next, we tested whether PirB-AP binds to cultured cortical neurons derived from brains of wild-type or β2m/Tap$^{-/-}$ mice. As with the MEFs, PirB-AP binds to neurons, and again this binding is reduced on neurons with low surface levels of MHCI (Fig. 2B), indicating that

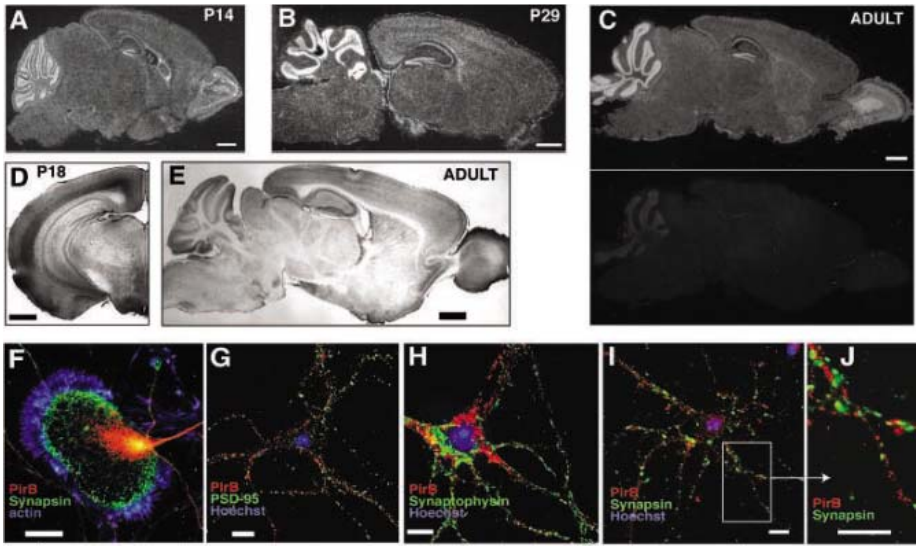
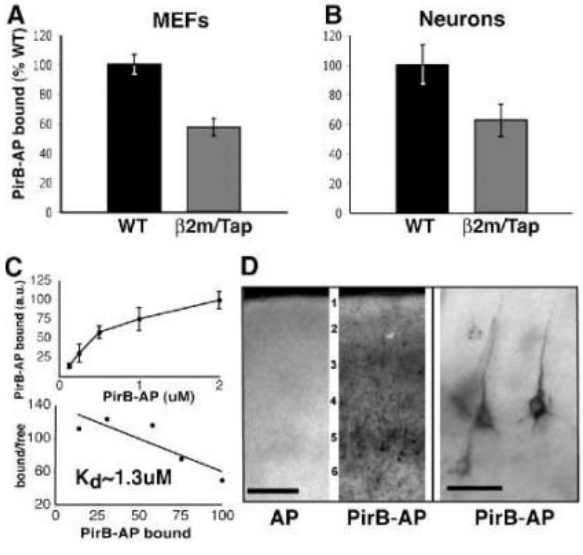


Fig. 1. PirB mRNA and protein are expressed in neurons. (**A** to **C**) ^{35}S labeled PirB-specific probes representing the 3′ region of PirB mRNA were used to detect PirB mRNA in sections from mouse brains of various ages. In situ hybridizations are shown in darkfield optics (silver grains appear white). (A) P14 sagittal section. (B) P29 sagittal section. (C) Adult sagittal section (top) plus sense control (bottom). (**D** to **J**) Immunohistochemistry using PirB-specific antibodies. (D) P18 coronal section and (E) adult sagittal section stained with the A20 antibody to PirB. Scale bars in (A) to (E), 1 mm. (F) Growth cone of a cortical neuron 3 days in vitro (DIV) immunostained with anti-PirB 1477 (red), phalloidin (blue), and anti-synapsin (green). (G) Cortical neuron 18 DIV stained with anti-PirB (red), postsynaptic marker postsynaptic density protein 95 (PSD-95) (green), and Hoechst (blue). (H) Cortical neuron 14 DIV stained with anti-PirB (red), presynaptic protein synaptophysin (green), and Hoechst (blue). (I) Cortical neuron 14 DIV stained with anti-PirB (red), presynaptic protein synapsin (green), and Hoechst (blue). (J) Higher magnification of (I). Scale bars in (F) to (J), 10 μm.

Fig. 2. Soluble PirB binds to neurons. Soluble PirB-AP binds to MEFs (**A**) and to cultured cortical neurons (**B**). Binding is dependent on surface expression of MHCI protein, given that binding is reduced significantly in cultures derived from β2m$^{-/-}$/Tap1$^{-/-}$ mice ($P <$ 0.01 for MEFs and $P = 0.03$ for neurons). a.u., arbitrary units. (**C**) Binding of PirB-AP to neurons is saturable (top). Scatchard analysis (bottom) predicts a dissociation constant of ~1.3 μM. (**D**) PirB-AP binds to pyramidal neurons in sections of cortex. Numbers indicate cortical layers. Scale bars, 250 μm (left and middle panels); 50 μm (right panel). Error bars in (A), (B), and (C) = 1 SEM.



soluble PirB binds to neurons in an MHCI-dependent manner. This binding is saturable (Fig. 2C), with relatively low affinity ($K_d \sim 1.3$ μM, Fig. 2C), consistent with observations of PirB-MHCI interactions in the immune system (*10*), as well as with other known MHCI-receptor interactions (*15*). In sections of the cerebral cortex, the PirB-AP fusion protein binds along the cell bodies and dendrites of cortical pyramidal neurons (Fig. 2D), consistent with the observation that panspecific antibodies to MHCI also immunostain the dendrites of neurons in the cortex and hippocampus (*4*).

Immunoprecipitation of PirB directly from the brain demonstrates that PirB exists primarily as a ~130-kD glycosylated protein (Fig. 3); this has also been observed in the immune system (*13*, *16*). Immunoprecipitation and Western blots were performed using different antibodies to ensure specificity. PirB can be detected in all brain regions at all ages tested (Fig. 3, A and B). We know that PirB from the brain is glycosylated because it is sensitive to deglycosylation by PNGaseF, which removes *N*-linked oligosaccharides (Fig. 3C); PirB is insensitive to EndoH, which cleaves a more restricted subset of oligosaccharides. Preparation of synaptosomes from mouse brain indicates that a substantial portion of PirB protein fractionates with synaptosomal plasma membranes but is distinct from the purified synaptic vesicle fraction (Fig. 3D), consistent with immunolocalization (Fig. 1, H to J) and suggesting that PirB may function at or near synapses.

The discovery of PirB expression in central nervous system neurons raises the question of neuronal PirB function. Therefore, we created a mutant mouse in which we removed four of the exons that encode the transmembrane domain and part of the PirB intracellular domain, rendering PirB unable to convey signals across the plasma membrane (fig. S1). We refer to the resulting mutant mouse, which is completely viable, and the mutant protein as PirBTM. To assess this mutation, PirB protein from the PirBTM mouse brain was examined. Figure 3E shows subcellular fractionation of wild-type and PirBTM brain, followed by immunoprecipitation of PirB and the shorter PirBTM mutant proteins, respectively. Wild-type PirB fractionates with both heavy and light membranes, with no signal detected in the soluble fraction. The mutant PirBTM protein is smaller and fractionates with light membranes and cytosolic fractions. Thus, the loss of the transmembrane domain has altered the solubility of PirB.

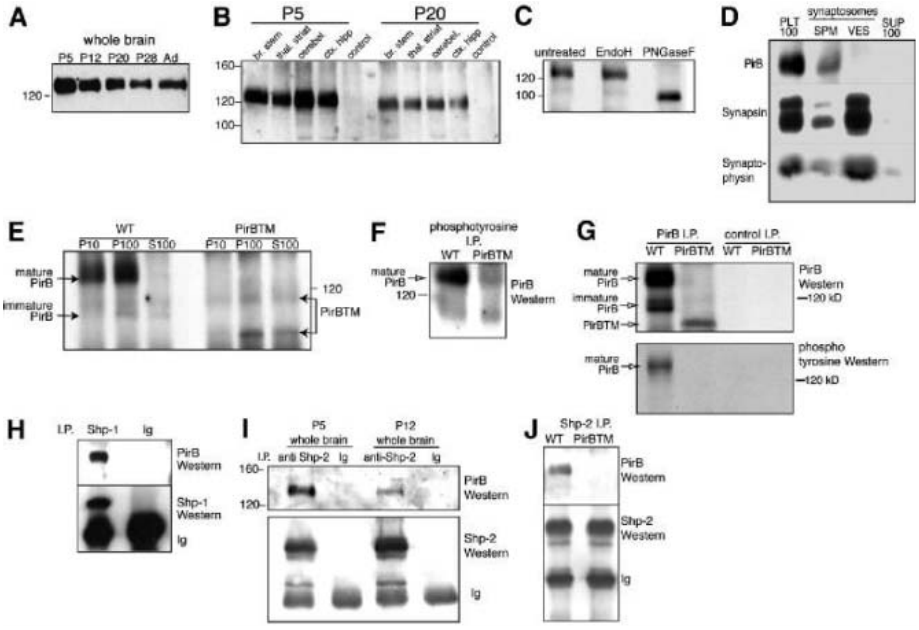


Fig. 3. PirB protein is expressed throughout the brain and forms complexes with Shp-1 and Shp-2. (**A**) PirB was immunoprecipitated using 6C1 monoclonal antibody and detected by Western blot using C19 polyclonal antibody from whole brain at P5, P12, P20, P28, or adult (Ad). (**B**) PirB protein immunoprecipitated from brain stem, thalamus and striatum (thal. striat), cerebellum, and cortex/hippocampus (ctx. hipp) derived from equal amounts of protein from P5 or P20 mouse brains. Control immunoprecipitation was nonspecific rat immunoglobulin in lysate from cerebellum. (**C**) PirB was immunoprecipitated from whole brain at P7 and treated with EndoH or PNGaseF glycosidases. (**D**) PirB was immunoprecipitated from synaptosomal fractions (*30*). PLT100, light membranes; SPM, synaptic plasma membranes; VES, synaptic vesicle fraction; SUP100, soluble fraction. Synapsin and Synaptophysin synaptic vesicle proteins were used as fractionation markers. (**E**) Subcellular fractionation from wild-type (WT) and PirBTM mouse brains. In wild-type mice, 130-kD mature PirB fractionates with heavy membranes (P10 fraction) and light membranes (P100 fraction), but none is detected in soluble S100 fraction. In contrast, mutant PirBTM is smaller and exhibits increased solubility; thus, a large proportion appears in the soluble (S100) fraction. (**F**) Anti-phosphotyrosine immunoprecipitation (I.P.) followed by anti-PirB Western blot reveals that PirB is phosphorylated, and no signal is detected in PirBTM mice. (**G**) Anti-PirB immunoprecipitation followed by anti-PirB Western blot, which is then stripped and probed for anti-phosphotyrosine. Only wild-type PirB is phosphorylated; no phosphorylated PirBTM is detected. (**H**) Anti–Shp-1 immunoprecipitation, followed by Shp-1 and PirB Western blots, demonstrates that PirB interacts with Shp-1 in brain. (**I**) Anti–Shp-2 immunoprecipitation followed by Shp-2 and PirB Western blots from P5 or P12 brains demonstrates a complex of PirB and Shp-2 in the brain. (**J**) The complex of PirBTM and Shp-2 is absent in PirBTM brains. Ig, immunoglobulin.

The cytoplasmic domain of PirB contains four immunoreceptor tyrosine-based inhibitory motifs (ITIMs). Phosphorylation of these sites is known to recruit Shp-1 and Shp-2 phosphatases to PirB, which in turn modulates signal transduction pathways in the immune system (*16*–*18*). When immunoprecipitation was performed with antibodies to phosphotyrosine from wild-type or PirBTM brains, followed by anti-PirB Western blot, we found that PirB was phosphorylated only in wild-type brains; no tyrosine-phosphorylated PirBTM protein was detected (Fig. 3F). This result was expected, given that PirBTM is not a transmembrane protein and thus is unable to engage ligand, which normally leads to phosphorylation (*10*, *11*). Similarly, when PirB or PirBTM is immunoprecipitated directly and analyzed for PirB (Fig. 3G, top panel), followed by phosphotyrosine Western blot (Fig. 3G, bottom), only wild-type PirB is phosphorylated. In immune cells, phosphorylated PirB recruits and signals primarily through Shp-1 and Shp-2 phosphatases (*8*, *16*); neuronal PirB also associates with these phosphatases (Fig. 3, H to J), suggesting that components of PirB-dependent signaling mechanisms are conserved between the immune and nervous systems. As expected, PirBTM does not coimmunoprecipitate with Shp-2 (Fig. 3J). Together, these observations demonstrate that the mutant PirBTM fails to transduce signals by means of phosphorylation of its remaining immunoreceptor tyrosine-based inhibitory motifs, which is the primary means by which PirB and other proteins of this class signal (*7*). Thus, we conclude that signaling through PirB in the brain of PirBTM mice is abrogated.

The search for neuronal MHCI receptors was inspired by the discovery that mice defective for MHCI surface expression have grossly normal brains but have specific abnormalities in synaptic connectivity and plasticity (*3*). In the visual system of $\beta 2m/Tap^{-/-}$ mice, the ipsilateral projection from the retina to the lateral geniculate nucleus (LGN) is larger than normal, consistent with a defect in developmental syn-

apse elimination. In the hippocampus, long-term potentiation is enhanced and long-term depression is absent, again consistent with a shift in synaptic plasticity toward strengthening at the expense of synaptic weakening (*3*). Initial examination of the PirBTM mouse also revealed no obvious phenotype. Gross brain histology was normal as assessed by Nissl staining (fig. S2A). In contrast to MHCI mutant mice, however, in PirBTM mice the gross pattern of connections from the retina to the LGN is indistinguishable from the wild-type mouse, with well-defined eye-specific domains (fig. S2B). This phenotypic difference likely reflects the fact that MHCI mutants are deficient in both the β2-microglobulin and the TAP1 genes, resulting in reduced surface expression of virtually all MHCI proteins [more than 50 in this large multigene family (*14*)]. Thus, MHCI mutant mice represent an extreme loss of function for all potential MHCI receptors expressed in the brain, including but not limited to PirB, as well as for possible receptor-independent MHCI functions such as those reported in the pheromone signaling system (*19*).

PirB protein is highly expressed in the cerebral cortex (Fig. 1). To examine whether PirB function is required in the cortex, the development of ocular dominance (OD) was assessed in the primary visual cortex of wild-type and PirBTM mice. The adult mouse visual cortex receives functional inputs from both eyes (Fig. 4A). Most of the visual cortex consists of a large monocular zone where neurons are visually driven exclusively by the contralateral eye. A more restricted region, the binocular zone (BZ), receives functional inputs from both ipsilateral and contralateral eyes (*20*, *21*). Dur-

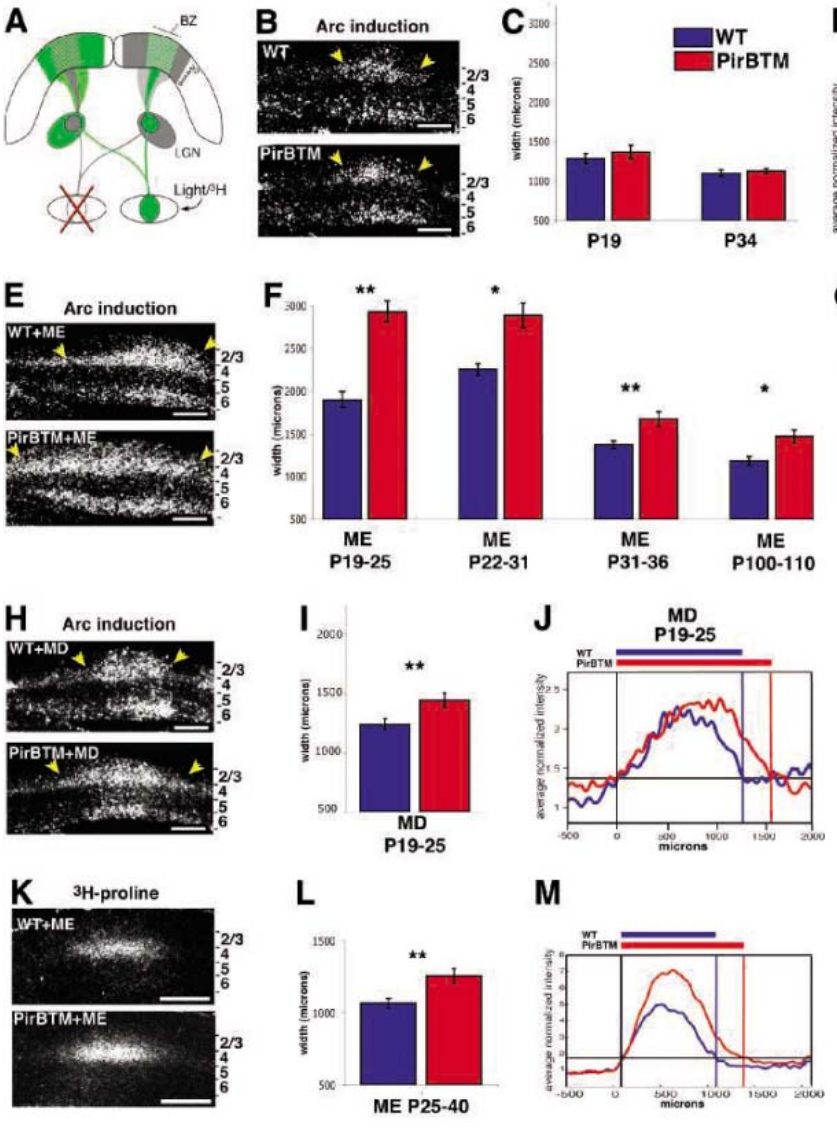


Fig. 4. Enhanced OD plasticity in visual cortex of PirBTM mice, as shown by Arc mRNA induction [(A) to (J)] and transneuronal autoradiography [(K) to (M)]. (**A**) Schematic of visual system showing connections from the retina to the LGN to the visual cortex. The small BZ receives visual inputs through the LGN from both eyes. (**B** to **D**) Developmental restriction of ipsilateral eye representation proceeds normally in PirBTM mice. (B) Arc mRNA induced by visual stimulation of P34 mice, detected by in situ hybridization in cortex ipsilateral to stimulated eye. Yellow arrowheads delineate zone of Arc induction, which corresponds to the BZ. In normally reared mice at P34, the width of Arc induction in wild-type or in PirBTM mice appears indistinguishable. (C) Histograms represent average widths of Arc induction in layer 4. P19: wild-type $n = 9$, PirBTM $n = 7$; P34: wild-type $n = 7$, PirBTM $n = 7$; three to four sections per animal. (D) Averaged line scans from all wild-type (blue) or PirBTM (red) sections at P34. Scans were made blind to genotype and were aligned at left border of BZ (black vertical line, left). Blue or red vertical line indicates right border of Arc induction. (**E** to **G**) OD plasticity after ME is enhanced in PirBTM mice. (E) OD plasticity after ME from P22 to P31 as assessed by Arc induction. The width of in situ hybridization pattern expanded after ME in wild-type mice [compare (E) top with (B) top]. Zone of Arc induction after ME is even more extensive in PirBTM mice [(E) bottom] than in wild-type mice [(E) top]. (F) Consistently enhanced expansion in width of Arc induction in PirBTM versus wild-type mice after periods of ME from P19 to P25, P22 to P31, P31 to P36, and P100 to P110. Histograms represent average width of Arc induction. ME from P19 to P25: wild-type $n = 5$, PirBTM $n = 9$; P22 to P31: wild-type $n = 6$, PirBTM $n = 6$; P31 to P36: wild-type $n = 9$, PirBTM $n = 9$; P100 to P110: wild-type $n = 5$, PirBTM $n = 5$; three to four sections per animal. (G) Averaged line scans of layer 4 Arc signal in all wild-type (blue) or PirBTM (red) sections after ME from P22 to P31. Scans aligned at left border of BZ (vertical line, left). Blue or red vertical lines indicate right border. The width is larger in PirBTM mice. (**H** to **J**) Enhanced OD plasticity after MD by eyelid suture from P19 to P25 in PirBTM mice: wild-type $n = 13$, PirBTM $n = 13$, three to four sections per animal. (**K** to **M**) Transneuronal autoradiography reveals an increase in width of anatomical connections between LGN neurons representing ipsilateral eye and layer 4 of cortex after ME (P25 to P40) in PirBTM mice. (K) Darkfield autoradiographs showing increased width of transneuronally transported radioactive label representing input from the ipsilateral eye in layer 4 of cortex in PirBTM (bottom) versus wild-type (top) mice. (L) Histograms are averages from all mice (wild-type $n = 7$, PirBTM $n = 6$). (M) Averaged line scans from all wild-type or PirBTM sections. Scans were aligned at left border of BZ (black vertical line, left). Blue or red vertical lines indicate width of thalamocortical projection in wild-type or PirBTM mice. Error bars in (C), (F), (I), and (L) = 1 SEM. $^*P < 0.05$; $^{**}P < 0.01$. Scale bars, 500 μm.

ing development however, neurons across a wider region of visual cortex receive functional inputs from the ipsilateral eye; by the fourth postnatal week, this region becomes restricted by activity-dependent mechanisms to the adult BZ (*22*–*24*).

The state of OD was assessed in the visual cortex by means of the activity-regulated immediate-early gene Arc. A brief (30-min) exposure of one eye to visual stimulation rapidly induces Arc mRNA exclusively in visual cortical neurons, revealing the extent and laminar distribution of cortical neurons functionally connected to the stimulated eye (*24*). In the hemisphere ipsilateral to the stimulated eye, a circumscribed zone of Arc induction (Fig. 4B) that coincides with the BZ (Fig. 4A) is present, allowing for quantitative high-resolution measurements of refinement and/or plasticity of the ipsilateral eye representation in the mouse (as well as the cat) visual cortex (*24*). This technique has the advantage, compared with other techniques, of not requiring anesthesia, which masks some forms of cortical plasticity (*25*). We compared the representation of the ipsilateral eye within the BZ of wild-type and PirBTM mice by means of Arc mRNA induction. All experiments and analyses were performed blind to genotype.

Stimulation of one eye induces Arc mRNA in cortical layers 2 to 4 and 6 [Arc is not expressed in layer 5 (*24*)]. In the hemisphere ipsilateral to the stimulated eye at P34, Arc induction is restricted to the BZ, revealing the adult pattern of OD (Fig. 4B) (*24*). The pattern of Arc induction in PirBTM mice at P34 is indistinguishable from that of wild-type mice at the same age (Fig. 4B). To quantify these observations, serial line scans were made through layer 4 across visual cortex, and the width of the zone of Arc induction was measured: The width in wild-type and PirBTM mice at P34 is identical (Fig. 4, C and D). It is conceivable that the BZ in P34 PirBTM mice arises from an earlier representation that is different from normal. However, the pattern of Arc induction at P19 is also indistinguishable between PirBTM and wild-type mice (Fig. 4C). In addition, the width of the zone of Arc induction is larger at P19 than at P34 in both genotypes (Fig. 4C). Together, these observations indicate that PirB function is not required for the normal developmental restriction of the ipsilateral eye representation within the BZ of visual cortex.

The OD of cortical neurons can be shifted readily by altering the relative amounts of activity between the two eyes; this is referred to as OD plasticity. The degree of OD plasticity is extensive during a critical period of development (*26*) and is far more limited at older ages (*23*–*25*, *27*–*29*). During the critical period, closing or removing one eye for several days shifts OD markedly toward the open, remaining eye. This shift in OD can be assessed directly by means of Arc mRNA induction in the visual cortex ipsilateral to the remaining eye; the Arc mRNA signal expands to occupy a wider-than-normal zone across layer 4 (Fig. 4E) (*24*), as well as other cortical layers. Unexpectedly, after a period of monocular enucleation (ME), OD plasticity in PirBTM mice was significantly enhanced compared with that of wild-type mice (Fig. 4, E to G) during the critical period. The difference held true for ME from P19 to P25, which overlaps the peak of the critical period; ME from P31 to P36, at the end of the critical period; and ME from P22 to P31, which spans the peak of OD plasticity. In addition, ME from P100 to P110, which is considered adult and well beyond the critical period, produced a greater expansion of Arc induction in PirBTM than in wild-type mice. At the ages examined, the width of Arc induction in layer 4 of PirBTM mice increased from 22% (P31 to P36; $P < 0.01$) to as much as 54% (P19 to P25; $P < 0.01$) of that in wild-type mice (Fig. 4F).

It is possible that the notable enhancement of OD plasticity in PirBTM mice is caused by the deleterious effects of eye removal itself, rather than by deprivation or visual experience. Therefore, the OD plasticity experiment was repeated using monocular deprivation (MD) by means of eyelid suture from P19 to P25. In wild-type animals, this technique is known to produce less plasticity than does ME (*24*); nevertheless, in PirBTM mice, the width of Arc induction in layer 4 was still 17% greater than in wild-type mice (Fig. 4, H to J). Together, these experiments imply that PirB limits OD plasticity induced by either ME or MD.

The induction of Arc mRNA in the BZ of the visual cortex is a functional measure of OD, delineating the area of cortex containing neurons responding to visual stimulation of the ipsilateral eye. Anatomically, the expanded Arc signal present in PirBTM visual cortex after a period of ME may be due to increased horizontal connectivity within the visual cortex, or it may be due to an increase in the spread of thalamocortical axon terminals from the LGN representing the ipsilateral eye (Fig. 4A), or both. To examine whether LGN axons increased their territory within layer 4, we performed ME on wild-type and PirBTM mice at P25; then, at P40, the LGN input from the remaining eye to layer 4 was assessed by means of transneuronal transport after an intraocular injection of 3H-proline (Fig. 4, K to M) (*30*). The transneuronally transported label within layer 4 of PirBTM mice is 18% wider than that of wild-type mice, indicating that at least part of the expansion in width of Arc mRNA induction observed in the PirBTM visual cortex is due to an anatomical increase in the area of layer 4 receiving inputs from LGN axons.

The enhanced OD plasticity seen in PirBTM mice both at the structural (thalamocortical axons) and at the functional levels (Arc induction in the postsynaptic cortical neurons) indicates that PirB normally functions to restrict the extent of cortical OD plasticity not only during but also after the critical period. In contrast, the normal developmental restriction of functional inputs from the ipsilateral eye to the BZ of the visual cortex is intact in PirBTM mice, as is the normal developmental remodeling of eye-specific projections from the retina to the LGN. Together, these experiments show that PirB is needed to restrict the ability of neural circuits to readjust synaptic connections in response to alterations in activity levels or balance of inputs. In the absence of PirB function, OD plasticity in visual cortex is enhanced at all ages, with a particularly strong effect seen during the critical period. Thus, in addition to mechanisms that enable synaptic plasticity, our results show that there are other mechanisms, such as those that involve PirB, that limit the extent of synaptic plasticity, which must then be determined by a balance of signaling in both pathways.

In the immune system, PirB acts through Shp-1 and Shp-2 phosphatases to inhibit signals that could lead to inappropriate and dangerous activation of immune cells against normal, healthy cells. PirB is thought to regulate cytoskeletal dynamics, cell motility, and adhesion, acting downstream of Src family kinases to modulate integrin signaling, Ca^{++} signaling, and kinase cascades (*18*, *31*, *32*). We report here that in neurons, PirB also recruits both Shp-1 and Shp-2. Thus, PirB may have analogous functions in restricting the response of neurons to activity-dependent or Ca^{++}-dependent signaling, thereby limiting aspects of synaptic plasticity. It is possible that cellular mechanisms that normally regulate and limit the selective strengthening or stabilization of synapses, the formation of new synapses, or even the outgrowth of new neurites after perturbations of sensory input are altered without functional PirB. This may result in exuberant and abnormal growth or strengthening of connections, as reflected in the increase seen here both in the width of thalamocortical connections representing the ipsilateral eye and in the width of Arc mRNA induction reflecting the functional activation of cortical neurons driven by the ipsilateral, open eye. In this context, important similarities emerge between the phenotypes of PirBTM mice and those observed previously in mice mutant for MHCI cell surface expression (*3*): In both strains of mutant mice, there is an enhancement of mechanisms that favor strengthening of synaptic connections.

It has long been known that there are critical periods during brain development when experience can rapidly alter circuits by changing synaptic connections, both by altering the structure of connections and by changing their strength functionally (*26*). Such periods are thought to be terminated by progressive developmental changes leading to an adult state in which plasticity is far more limited if it occurs at all. Here, we show that both during and after

the critical period, the extent of plasticity is actively constrained by PirB. In the immune system, PirB is known to regulate integrin-dependent cytoskeletal dynamics (*31*, *33*), and integrin activation in neurons can affect several aspects of synaptic function and plasticity (*34*–*37*). In the visual system, studies show that the tissue plasminogen activator/plasmin extracellular proteolytic cascade is essential for structural plasticity in the visual cortex (*38*) and cleaves extracellular proteins including the integrin ligands laminin and fibronectin (*39*). Thus, PirB may restrict neuronal plasticity by affecting the ability of activated integrins to engage the neuronal cytoskeleton. Upon closure of the critical period, factors thought to constrain plasticity include Nogo Receptor (*40*) and extracellular matrix molecules chondroitin sulfate proteoglycans (*41*). How PirB function interacts with these factors is not known, but our experiments demonstrate a role for PirB in limiting the extent of synaptic plasticity in the visual cortex and may provide insight into mechanisms that are needed to stabilize neural circuits.

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Supporting Online Material

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The Garvan Institute of Medical Research is one of Australia's pre-eminent biomedical research Institutes. The research of the Institute is carried out within seven major research disciplines - Cancer, Diabetes & Obesity, Immunology & Inflammation, Bone & Mineral and Neuroscience Research Programs and the Autoimmunity and Pituitary Research Units.

The goal of the Neuroscience Research Program (**http://www.garvan.org.au**) is to increase our understanding of the neuronal systems involved in a variety of neurological disorders. Our scientists use molecular, electrophysiological, physiological and anatomical approaches, including site-directed mutagenesis, conditional knockout technology, stem cell differentiation, patch clamp recording and confocal microscopy to address these questions.

Garvan Institute is inviting applications from internationally recognised scientists for the position of Chair in Neuroscience supported by the Curran Foundation. This is a full-time, five year renewable position. The successful applicant will be expected to direct internationally competitive research in the area of **hearing research** that complements and collaborates with existing groups within the Neuroscience Program and other programs in the Institute, and in so doing further develop the Institute as an international Centre of Excellence in Neuroscience Research.

Applicants should hold a PhD, or equivalent postgraduate qualifications, and an international record of outstanding scientific achievement in any area of neuroscience research of direct relevance to hearing. Excellent interpersonal, written and oral communication skills, in conjunction with previous management experience are essential.

An attractive remuneration package will be offered to the successful applicant, commensurate with qualifications and experience.

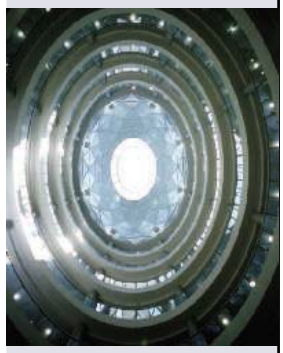
Garvan reserves the right to fill the position by invitation or not fill the position.

For further information please refer to the Garvan website: www.garvan.org.au. Enquiries regarding this position should be directed to Professor Herbert Herzog, Director Neuroscience Research Program, Tel +61 2 9295 8296.

Applications should include: Curriculum Vitae, proposed research objectives and the contact details of three referees, and be forwarded to:
Human Resources
Garvan Institute of Medical Research
384 Victoria Street, Darlinghurst NSW 2010, Australia
Facsimile +61 2 9295 8101 Email: hr@garvan.org.au

An information package is available, please contact the Human Resources Department on hr@garvan.org.au to obtain a copy.

Curran Foundation Chair in Neuroscience Research



Applications close Friday, 27th October 2006.

GARVAN INSTITUTE

International Careers Report: Australia

Punching above Its Weight

Despite its small population and relative remoteness, Australia occupies a powerful position in global science in general and life science in particular. The ingredients of its success: a strong research infrastructure, solid governmental support, effective collaboration among academic, industrial, and governmental institutions, and an ambience that attracts scientists from around the world. BY PETER GWYNNE

AusBiotech
http://ausbiotech.com

Australian Research Council
http://www.arc.gov.au

Australian Stem Cell Centre
http://www.stemcellcentre.edu.au

Centre for Immunology and Cancer Research
http://www.cicr.uq.edu.au

Department of Education, Science and Training
http://www.dest.gov.au

Garvan Institute of Medical Research
http://www.garvan.org.au

Griffith University
http://www.griffith.edu.au

Institute for Molecular Bioscience
http://www.imb.uq.edu.au

Kelly Scientific Resources
http://www.kellyservices.com

University of Western Australia
http://www.uwa.edu.au

Modern Australia offers the world much more than first-class cricket and shrimps on the barbie. With a population of just 20 million, this nation has emerged as the top ranking location for biotechnology in the Asia-Pacific region and as a major global player in both physical sciences and academic and industrial life science. The country has excellent research infrastructure for life science and physical science, a powerful emphasis on collaborative projects and shared facilities, strong government funding of research, and an openness to scientists of all types – and particularly life scientists – from around the world.

"Australia is absolutely punching above its weight in life science," says Anna Lavelle, CEO of AusBiotech, Ltd. "It has 0.3 percent of the global population, 2.8 percent of peer-reviewed papers, and significant global patents. Australia is No. 6 in the world in terms of companies and activity in biotechnology."

Stephen Livesey, CEO of the Australian Stem Cell Centre, outlines why the lucky country has proved so successful. "Excellent research facilities, innovative scientists, a can do attitude, and a strong but flexible regulatory regime have made Australia a life sciences powerhouse," he explains.

The authorities plan to keep it that way. "The Australian government has made a long-term commitment to building a world-class science and innovation system through CONTINUED »

International Careers Report: Australia

our 10-year A$8.3 billion (US$6.3 billion) Backing Australia's Ability and Backing Australia's Future packages," says Julie Bishop, Minister for Education, Science and Training. "It has also introduced national research priorities to guide Australia's effort in areas of economic, social, and environmental importance."


JULIE BISHOP

Collaborative Projects

That guidance includes strong support for collaborative projects. "The National Health and Medical Research Council has set up a structure that encourages people to come together in teams," says Brandon Wainwright, professor of molecular genetics and director of the University of Queensland's Institute for Molecular Bioscience. The Commonwealth Scientific and Industrial Research Organization (CSIRO), the national agency that supports research and development, funds cooperative work through its Flagship Collaborative Research Programme.

That program has helped to stimulate another ambitious project. "The federal government has tried to encourage formation of Cooperative Research Centres – combinations of a university-based research group with a particular industry and perhaps a connection to CSIRO or similar institution," says George Stewart, dean of the faculty of life sciences at the University of Western Australia. Those centers, adds John Shine, executive director of the Garvan Institute of Medical Research, "have been very effective in facilitating increased interaction between academic, industrial, and governmental research."

Life science has received particular benefit. "The cooperative research centers have fostered the growth of strong intersectoral collaborations across the field of biosciences," observes Ian Frazer, director of the University of Queensland's Centre for Immunology and Cancer Research, and Australian of the Year 2006.

The centers focus on more than basic research. "We're not only about discovery science but also translation science from which we can derive benefits for mankind," explains Mark von Itzstein, executive director of the Institute for Glycomics and professor of medicinal chemistry and federation fellow at Griffith University. Researchers and government granting organizations agree on the need to push research out of the lab. "We believe it's a moral imperative to get the results of our research back to the people who paid for it – the taxpayers," Wainwright asserts. "If we have a finding that we can apply, we'll do that. The government has an emphasis on outcomes." Peter Høj, CEO of the Australian Research Council, points out the effect of that dynamic. "Even in nondirected research programs," he says, "researchers tend to end up in areas important for the economy."


IAN FRAZER

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PETER HØJ

Beyond the Borders

Collaborative efforts extend beyond Australia's borders. "Schemes like the government's International Science Linkage program and the Australian Research Council's Linkages International program provide Australian researchers with access to international expertise, facilities, and projects," Bishop says. "We are building scientific networks and alliances of a depth and scale that allow us to share knowledge across borders and sectors." Industry takes a similar approach. "More than 70 percent of the 339 Australian life science alliances announced during 2005 were with organizations outside the country," Livesey says.

Australia has plenty of natural resources to offer overseas collaborators. Since 1993, for example, Europe-based pharma AstraZeneca has invested more than A$100 million (US$76 million) in seeking new drug compounds in Australian flora and fauna. The project, in cooperation with Griffith University, has identified more than 700 bioactive compounds with druggable potential.

Overseas organizations have also cast envious eyes on another Australian resource – its young scientists. "We're missing people coming through into postgraduate work here," Stewart says. "A number of our top graduates go overseas to Europe or North America." Høj emphasizes that point. "There's a global talent war going on," he says. "Australia needs to have a research system good enough to ensure that its scientists aren't sucked off permanently to other countries."

In recent years, however, Australia has begun to turn a brain drain into a brain gain. Increasing numbers of Australian scientists who go abroad for their postdoctoral research return to take up senior positions. Immigrants have joined them. "Recruitment from overseas has been rapidly growing in recent years, due to increased funding and more awareness of both Australian science and lifestyle," Shine reports. "Approximately half of our postdoctoral fellows at the Garvan Institute are from overseas."

It's hardly surprising that overseas organizations seek out Australian life scientists. The country has a proud history of achievement in the field. "We have five Nobel laureates in physiology or medicine," von Itzstein points out. "That demonstrates quite clearly that there is a very strong life science base within this country." Current laureates in residence include Peter Doherty, who shared the 1996 prize for discovering how the body's immune system recognizes virus-infected cells, and Barry Marshall and Robin Warren, who received the award last year for their discovery that the bacterium Heliobacter pylori causes stomach ulcers and gastritis. CONTINUED »

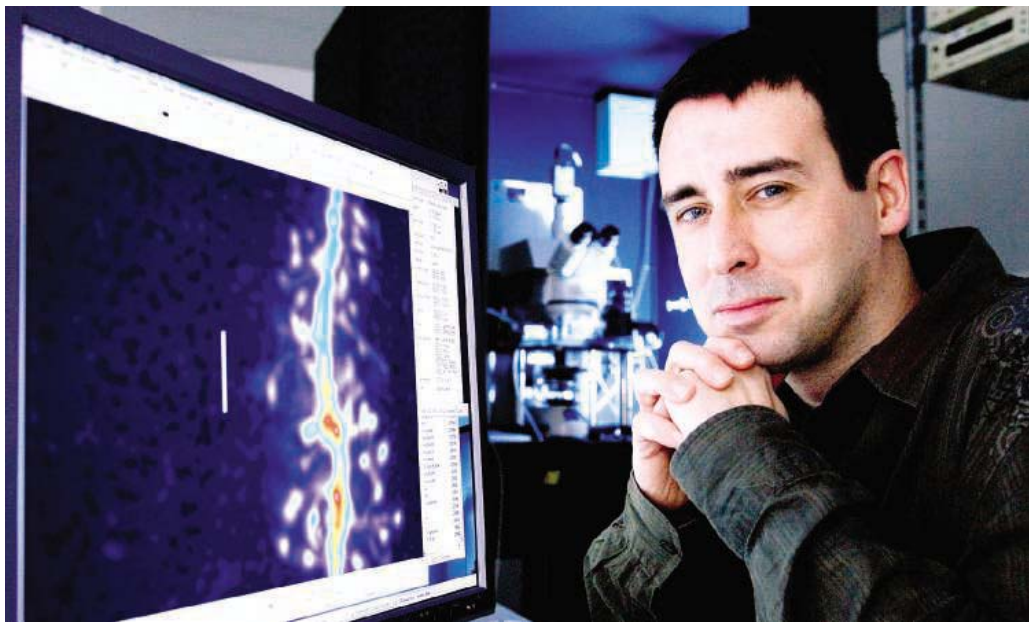
Join one of the world's most exciting neuroscience research teams at the Queensland Brain Institute in Australia

Neuroscience is entering an era of accelerated discovery, promising a new understanding of the mechanisms that regulate brain function and applying this to developing new therapeutics for brain diseases. With this aim, The Queensland Brain Institute (QBI) – part of The University of Queensland – is recruiting up to six new Faculty in the areas of cellular and synaptic plasticity, visual, cognitive and computational neurosciences. The foundation Director of QBI, Professor Perry Bartlett FAA, heads a dynamic Faculty of senior research neuroscientists which includes Pankaj Sah, Mandyam Srinivasan FAA, FRS, Brent Reynolds, Jason Mattingley, Geoffery Goodhill, Linda Richards and Helen Cooper.

Established in 2003, QBI is home to world-leading research in cellular and synaptic plasticity. Leadership direction builds on the key discovery made by several Faculty members in 1992 that the adult brain contains stem cells capable of producing new neurons. QBI is exploring ways to stimulate the production of new functional nerve cells to overcome diseases such as dementia, stroke and motor neuron disease and to promote optimal brain function.

QBI's research focus is on discovering and understanding the molecular and physiological regulation of brain function. Specifically, QBI targets brain plasticity with six main research themes, all of which will be expanded, in terms of infrastructure and research capacity, over the next five years:

- Cellular plasticity
- Synaptic plasticity
- Cognitive and behavioural neuroscience
- Visual neuroscience
- Mental and neurological disorders
- Computational neuroscience



Above: QBI Research Fellow, Dr John Power

World-class facilities

The Institute's potential to make key advances will be further progressed by the development of a $64 million facility, fitted with state-of-the-art research equipment, and have the capacity to accommodate some 250 scientists and support staff.

Scheduled for completion in July 2007, the new complex will see the Institute evolve into one of the finest neuroscience research centres in the world. QBI's scientific team will also develop leadership in core technology, especially in advanced imaging. The Institute has already installed a16.4Tesla animal MRI and 4T human MRI. In addition, QBI has the world's largest cell-sorting facility dedicated to neuroscience. The new Institute will also have extensive capabilities in animal and human behavioural testing and has developed strong interdisciplinary teams in the area of applying nanotechnology to neuroscience.

Asia–Pacific focus

By developing a highly collaborative network based on 'real' scientific exchanges and joint programs, QBI intends to become a focus for the Asia–Pacific neuroscience community.

Neuroscientists at all levels – up to full professor – are encouraged to apply.

Enquiries to:

Professor Perry Bartlett
p.bartlett@uq.edu.au
www.qbi.uq.edu.au

The University Of Queensland AUSTRALIA | qbi queensland brain institute

International Careers Report: Australia


ANNA LAVELLE

Abundance of Resources

Complementing the people, the country has an abundance of physical resources. "It's hard to think of something we don't have," Wainwright says. "There's a synchrotron that's almost finished and other devices at the really heavy end of life science. We have a national genomics facility. We have extensive facilities in microscopy and cryo-electron microscopy. At the Institute for Molecular Bioscience we have just taken delivery of a 900 megahertz NMR. We're extremely well equipped for large science research, from synchrotrons to animal houses." Von Itzstein of the Institute for Glycomics provides his own take. "In terms of the infrastructure and mentoring scientists, we are exceptional," he asserts. "We have more than a dozen well-established research institutes operating in life and multidisciplinary science."

The country also possesses human and mechanical expertise in a wide range of life science disciplines. "The high quality of Australian research is particularly apparent in fields such as clinical medicine, biology, immunology, environmental sciences, and space science," Bishop says. Stewart sees an evolution in specialties. "Traditionally, Australia always had a strong plant science capability, largely because of its origin in agricultural research," he says. "More recently it's become more focused on molecular plant biotechnology, as well as immunology and genetics. The country has targeted niche areas rather than a broad front." That approach provides a moving target. "Immunology is an ongoing strength," Wainwright says. "That's been knocked on through isolation of new growth factors. There's a new cancer vaccine center here in Brisbane; we have a lot of strength in vaccine research. And we're seeing an emerging strength in molecular cell biology."

Recent additions to the Australian life science portfolio include research on stem cells. "Australia has cutting edge capabilities in stem cell research and is home to world-class scientists who undertake research and development on embryonic and adult stem cells under a clear and transparent national legislative framework," Livesey explains. "Our stem cell research has a distinct competitive advantage in the area of hematology; Australian groups have been able to develop primitive blood cells from embryonic cells. The Australian Stem Cell Centre is investing significant resources in pursuing this interest with a view to producing a manufactured, safe, and reliable supply of blood products from stem cells."

Industrial Strength

That leads inevitably to industry, which can boast its own strength in life science. "Australia is home to a thriving network of 420 companies, up from 190 in 2001, whose core business is biotechnology," Livesey points out. "Of those, 48 percent are involved in human therapeutics, 16 percent in agricultural biotech, and 14 percent in diagnostics. Another 612 companies focus on medical devices." The sector contains globally recognized performers. "CSL is the world No. 1 in blood fractionation," AusBiotech's Lavelle says. "We have two very successful global medical device companies in Cochlear and Resmed."


STEPHEN LIVESEY

Lavelle pinpoints one disadvantage. "We are very strong in drug discovery, but we don't have a strong local pharmaceutical industry," she says. "That hinders drug development."

It also points out, by omission, the value of collaboration among universities, government, and industry in stimulating life science research and development. "Building the connections between science and industry is a key theme in the Australian government's approach to science and innovation," Education, Science and Technology minister Bishop says. "Collaboration between researchers is already the dominant pattern for research activities in Australia, and this is complemented by a focus on maximizing results and improving the commercialization of new ideas." As Shine of the Garvan Institute sees it, "The process is leading to an evolving research culture more accepting of, and proactively seeking, support from industry."

Stewart at the University of Western Australia has first-hand experience of that culture. "The federal government is now encouraging more informal networks that link universities and industry," he says. "For example, we're a node of the nano major research facility. We have certain of the microscope facilities here; others are at Queensland. There's a group of four universities involved that get state-of-the-art equipment through government grants to work on nanotechnology."

Does Australia produce enough scientists to satisfy the requirements of its academic and industrial research enterprises? CONTINUED »

Opportunities for Overseas Scientists


ANNE SABINE

As Australia's commitment to converting research into products continues, its institutions must work harder to recruit technical personnel. "The overall lack of qualified scientists has meant that companies need to cast their recruiting nets wider in order to find the right candidates," says Anne Sabine, director, Australia, for placement firm Kelly Scientific Resources. "The greatest demand is for cell biologists, molecular biologists, and protein chemists. In some cases we cannot keep up with the demand." Industry in particular finds a lack of qualified graduates from Australian universities. As a result, Sabine continues, "many positions are now being filled by cross-border recruitment efforts."

International Careers Report: Australia

"We are currently in balance, in the life sciences," Høj says. But he warns that science has declining appeal. "There is a concern that science as a career is not seen as attractive to young school leavers," he says.


BRANDON WAINWRIGHT

Rite of Passage

For young Australians who commit to science, another issue stems from the fact that many undertake what has become virtually a rite of passage. "Australian Ph.D.s are very much in demand in overseas laboratories," explains the Institute for Molecular Bioscience's Wainwright. "And a lot of Australians tend to go abroad. It's seen as a backwards career move to stay in the country. So we don't hold on to that high a number of top Australian graduates."

Stewart agrees. "It's getting more and more competitive to get first class postgraduate students," he says. "A number of our top graduates go overseas, to Europe or North America."

Von Itzstein of the Institute of Glycomics points out that Australian organizations' demands on their scientists have evolved. "There is a fine set of life scientists coming through our excellent research and teaching institutions," he says. "But like other countries, we need to educate our scientists in a more multidisciplinary way. You can no longer be simply a fine biologist; you need to be integrated."

To find integrated scientists, Australian companies, universities, and research centers must recognize the need to seek help on a worldwide basis. "We see research as being conducted in a global village, and recruit internationally on merit grounds," Frazer says. "It's of note that we have more senior scientists in the Centre for Immunology and Cancer Research from overseas than from Australia."

That's a common experience. "Of the postdoctoral fellows in this institute, about half are from overseas," Wainwright notes. "More than 25 countries are represented: Asian countries and a lot of South Americans, Canadians, and Europeans. We seem to have a bit of a pipeline of European undergraduates coming through for six months or so. But we're underrepresented from the United States." Recruitment from abroad has also blossomed at the Garvan Institute. "Approximately 50 percent of our postdoctoral fellows are from overseas," Shine says. "We have very few from the U.S.A., some from Canada. A majority comes from Europe – France, the United Kingdom, Spain, Germany, and Switzerland. And a growing number come from Asia, especially China."

Different Skill Sets

Overseas recruits carry with them more than different accents and languages. "We have extensive recruitment from overseas, because it brings in different skill sets and further consolidates international collaborations," von Itzstein says. "We get scientists from Asia, Europe, Canada, and the United States. It's an absolute joy to have an internationally based organization that's in touch with the world."

The federal government has begun to play its part in "ensuring that we continue to benefit from 'brain circulation'," as Bishop puts it. "The Australian Research Council's Federation Fellowships scheme, for example, encourages Australian researchers working overseas to bring their skills back into Australian science, while scholarships offered under the Endeavour Programme allow high achieving students from around the world to undertake study or research in Australia."

The Australian Stem Cell Centre offers its own incentives. "Our Premier Scholarships are aimed at bright students who are preparing to undertake a Ph.D. in the area of stem cell science," Livesey says. "Currently the Centre funds and manages 12 Ph.D. students. And to augment the long lead time necessary in developing Ph.D. students, we are actively recruiting young scientists to move to Australia. They have arrived from North America, the Southeast Asian region, and Europe."

What attractions beyond first class science does Australia offer to students and career scientists from overseas? "One of the huge advantages is the lifestyle here," AusBiotech's Lavelle says. "Australians work very hard, but have a nice country – mild weather and a stable, law abiding environment. The educational standard is very high. We have a good, affordable medical system. People with families find all that very attractive. Once they get here, settling in is a simple process."

The country has some downsides. "People sometimes find that the number of specialized roles is fewer than in a much larger country," Lavelle says. More important is what many term "the tyranny of distance." The country lies far from the major global centers of science. "The greatest advantage is also probably the greatest disadvantage," Livesey says. "Australia may be the most livable and beautiful place to live and practice science, but it is also a long way from Europe and North America."


JOHN SHINE

Unencumbered Travel

However, administrators work hard to overcome that problem and any others that newcomers might experience. "We provide travel money for Ph.D. students and postdocs so that they are not encumbered from traveling to their favorite overseas meetings," Wainwright says. And at the Center for Immunology and Cancer Research, Frazer adds, "We take the belief that, at whatever level we recruit, we need to provide a sufficient startup package to ensure that their research productivity is not compromised by the move to our center. This includes salary and research support packages over three to five years from appointment."

For foreign scientists convinced that Australia is a golden destination, Lavelle adds one extra piece of advice. "Remember," she says, "to bring your sunscreen."

A former science editor of Newsweek, *Peter Gwynne (pgwynne767@aol.com) covers science and technology from his base on Cape Cod, Massachusetts, U.S.A.*

Professor of Climate Modelling

Monash University, Melbourne, Australia

Monash University seeks an outstanding scholar for appointment as a Professor of Climate Modelling in the School of Mathematical Sciences to provide leadership in research and teaching in atmospheric science and to foster a strong collaboration between Monash University, the Bureau of Meteorology and CSIRO. The appointment is expected to lead Monash University's involvement with the Australian Community Climate Earth Systems Simulator.

Excellence in research and education and great diversity in location, culture and people distinguish Monash as a leading Australian and proudly international university. With campuses in Australia, Malaysia and South Africa and centres in the UK and Italy, it provides exciting international research and education opportunities. National and international students benefit from extensive curriculum choices offered by Monash's ten faculties in the sciences, professions and humanities.

The appointment will be made in the School of Mathematical Sciences, which has a strong research group in atmospheric science. Comprising 29 academic staff, the School of Mathematical Sciences in the Faculty of Science has research interests in algebra and discrete mathematics, analysis and geometry, applied statistics and probability, astrophysics and general relativity, meteorology and geophysical fluid dynamics, and computational mathematics.

The successful candidate will have: a research doctorate in a relevant discipline, an outstanding international reputation for research in an area of atmospheric science relevant to climate modelling; demonstrated success in establishing and leading successful research teams and innovative research programs; a proven capacity to attract competitive research funding; a record of successful supervision of postgraduate students; and demonstrated skills in management and development of staff.

Appointment will be on a continuing basis.

Remuneration: Professorial salary: $A121,459 per annum, plus superannuation. Relocation travel and removal allowances and salary packaging are available.

Selection documentation may be accessed electronically on the world wide web: http://www.adm.monash.edu/sss/employment/senior

Inquiries of an academic nature may be directed to Professor Michael Reeder, School of Mathematical Sciences, telephone +61 3 9905 4464, facsimile +61 3 9905 4403, email michael.reeder@sci.monash.edu.au

Applications that must specifically address all of the selection criteria should reach Ms Bronwen Meredith, Manager, Senior Academic Appointments (Advertised), Monash University, Victoria 3800, Australia, no later than Friday 20 October 2006.

Inquiries regarding the application process may be directed to Ms Meredith, telephone +61 3 9905 6193, facsimile +61 3 9905 6016, email bronwen.meredith@adm.monash.edu.au

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■ INSTITUTE OF MOLECULAR BIOSCIENCE (IMB)

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The IMB is organised into four divisions which together conduct cutting edge research into the genetic, cellular and molecular basis of normal and abnormal mammalian development and variation. The Group Leader will demonstrate outstanding achievement in molecular bioscience that fits within the strategic research directions for the IMB. The appointee will be expected to develop a visionary program that applies high-throughput proteomic approaches to understanding mammalian biological systems. This research program will complement the expertise of the senior IMB researchers in computational, structural, chemical, cellular and developmental biology. The appointee will be expected to provide academic oversight to senior staff running the extensive proteomics and mass spectrometry infrastructure of the IMB.

The Queensland Bioscience Precinct offers an outstanding research environment in systems biology and is equipped to a high standard in all aspects of high performance computing/bioinformatics, structural biology, molecular cell biology and genomics. Brisbane offers one of the world's best lifestyles, with a mild subtropical climate.

Remuneration: The appointment may be at any level between Senior Research Fellow (Research Academic Level C) and Professor/Senior Principal Research Fellow (Research Academic Level E) (approx salary range AUD$91,303 – 105,278 (Level C); AUD$109,936 – $121,116 p.a. (Level D); $141,613 p.a. (Level E), which includes 17% employer superannuation). Appointment will be at a level commensurate with the successful applicant's experience and achievement and will be accompanied by an appropriate initial support package.

Contact: To obtain the position description please contact Barb Clyde, telephone +61-7-3346-2121 or email b.clyde@imb.uq.edu.au. Informal enquiries about this position are most welcome. Please contact either Professor Brandon Wainwright, Director IMB, email b.wainwright@imb.uq.edu.au or telephone +61-7-3346-2110; or Professor John Hancock, email j.hancock@imb.uq.edu.au or telephone +61-7-3346-2033.

Reference No: 3008127.

Applications close: 23 October 2006.

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Look into INRO
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The National Institute of Allergy and Infectious Diseases (NIAID), Division of Intramural Research (DIR), is seeking applicants for its Intramural NIAID Research Opportunities (INRO) program. INRO, currently in its fifth year, is a 5-day exploratory program intended for underrepresented minority students (i.e., American Indians or Alaska Natives, Native Hawaiians or other Pacific Islanders, blacks or African Americans, and Hispanics or Latinos) who are interested in a research career in the areas of allergy, immunology, and infectious diseases.

This 5-day program will be held February 4–8, 2007, at the National Institutes of Health in Bethesda, Maryland. Applicants to this highly competitive program should be undergraduate juniors or seniors interested in postbaccalaureate biomedical research, doctoral candidates seeking a postdoctoral training position, and first-year medical students contemplating a year-off research position.

Students selected to attend the INRO 2007 program will:

• Learn about the Institute and about training opportunities available at NIAID • Listen to scientific lectures by world-renowned NIAID researchers • Tour Institute laboratories • Interview with principal investigators for potential training positions

The INRO program will pay all expenses for travel, hotel accommodations, and meals. Eligible students include U.S. citizens or legal U.S. residents who belong to a minority population that is underrepresented in the sciences. Only students with strong academic standing will be considered.

For more details about student eligibility, highlights of last year's INRO program agenda, student testimonials from past programs, and detailed information about the application process, students may visit our Web site at www.niaid.nih.gov/labs/training/inro. Completed applications MUST be received by **October 15, 2006**.

For additional information, contact **Wendy J. Fibison, Ph.D., Associate Director, DIR, NIAID, wfibison@niaid.nih.gov.**

NATIONAL CANCER INSTITUTE

SENIOR LEVEL RESEARCH FELLOW
Chromosome Segregation and Cell Cycle Checkpoint Control in *S. cerevisiae*

With nation-wide responsibility for improving the health and well being of all Americans, the Department of Health and Human Services (DHHS) oversees the biomedical research programs of the National Institutes of Health (NIH) and those of NIH's research Institutes.

The Genetics Branch (GB), Center for Cancer Research (CCR), National Cancer Institute (NCI) is recruiting for a Senior Level Research Fellow. Using *S. cerevisiae* as a model system, research in our laboratory is focused on three major areas: 1) Mechanism of faithful chromosome transmission, 2) Cell cycle and checkpoint regulation, and 3) Identification and characterization of previously non-annotated small open reading frames. The high degree of conservation between yeast and human genes makes *S. cerevisiae* an attractive model system for elucidating how failures in chromosome transmission may give rise to diseases such as cancer in humans. Results from our laboratory and those of others have shown functional complementation of yeast mutant phenotypes by human homologs. In addition to conventional approaches we employ genome wide-approaches such as microarrays and high throughout robotics that facilitates genetic screens, finding drug targets and studying the expression of genes from human in yeast. The Research Fellow position is a senior level position for candidates with post-doctoral experience. NIH provides interactions with almost 30 yeast laboratories in the local area. Salary range is 42K to 77K commensurate with experience. Candidates must have a Ph.D. and/or M.D and preferably experience in yeast genetics. Send CV, a statement of research interests and names of three references to: **Munira A. Basrai, Ph. D., Senior Investigator, Genetics Branch, National Cancer Inst./NIH, Navy Med. Ctr., Bldg. 8, Rm. 5101, 8901 Wisconsin Avenue, Bethesda, MD 20889. Email: basraim@nih.gov**.

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Department of Health and Human Services
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National Institute of Allergy and Infectious Diseases

With nation-wide responsibility for improving the health and well being of all Americans, the Department of Health and Human Services oversees the biomedical research programs of the National Institutes of Health (NIH) and those of NIH's research Institutes.

The National Institute of Allergy and Infectious Diseases (NIAID), a major research component of the NIH and the Department of Health and Human Services, is recruiting for a Tenure/Tenure Track position in the Laboratory of Host Defenses (LHD). The LHD studies immune functions essential for host defense against infection (inherited immune deficiencies) and those required for immune homeostasis (autoimmunity associated with excessive inflammation). The LHD seeks an M.D. or M.D., Ph.D. physician scientist to develop an independent translational research program related to the genetic basis, pathophysiology, diagnosis and treatment of autoimmune diseases associated with excessive inflammation. An emphasis on clinical aspects of innate immunity including phagocytic cells, natural killer cells, dendritic cells and other antigen presenting cells, toll-like receptors or other pattern recognition receptors in its interface with acquired immunity is desirable. The applicant should have a strong track record of basic research of the genetic basis of disease and alterations in signaling pathways responsible for immune dysregulation. The applicant must possess expertise and experience in the design and conduct of diagnostic and therapeutic clinical trials studying and treating autoimmune diseases. Strong clinical credentials in a specialty area relevant to the proposed translational research program (relevant specialties include but are not limited to rheumatology, pulmonary diseases, hematology, immunology or infectious diseases) are required. The program of study proposed by the applicant must include both laboratory components and the conduct of clinical protocols to assess new diagnostic and therapeutic modalities to diagnose and treat autoimmunity associated with excessive inflammation. Applicants particularly suitable for this program are those who have knowledge and experience in the development and clinical application of novel biological agents including chemokines, soluble chemokine receptors, adenosine receptor agonists, monoclonal antibodies, cellular therapies including transplantation or gene therapy to correct the abnormalities in immunity, that achieve immune tolerance or to reduce abnormal inflammation.

The applicant must provide evidence in the submitted materials that the applicant has a current license to practice medicine in one of the states of the United States or must have all the credentials required by the State of Maryland for licensing to allow the practice of medicine. These credentials must include but are not limited to having a Doctor of Medicine or Doctor of Osteopathy degree from an accredited school in the U.S. or Canada, or a Doctor of Medicine or equivalent degree from a foreign medical school that provided education and medical knowledge substantially equivalent to accredited schools in the U.S. as demonstrated by permanent certification by the Educational Commission for Foreign Medical Graduates (ECFMG).

To be considered for this position, you will need to submit a curriculum vitae, bibliography, three (3) letters of reference, a detailed statement of research interests, and a hardcopy of selected publications to **Thomas A. Fleisher, MD, Chairperson, NIAID Search Committee, c/o Ms. Anissa N. Hunter, DIR Committee Coordinator, Reference Ad #009, 10 Center Drive MSC 1356, Building 10, Rm. 4A26, Bethesda, Maryland 20892-1356**. Completed applications **MUST** be received by **Thursday, November 15, 2006**. For additional information on this position, and for instructions on submitting your application, please see our website at: **www.niaid.nih.gov**.

Positions @ NIH

The National Institutes of Health

Chief, Laboratory of Virology
National Institute of Allergy and Infectious Diseases
National Institutes of Health

The National Institute of Allergy & Infectious Diseases (NIAID), Division of Intramural Research (DIR) is seeking an outstanding individual to head the newly established Laboratory of Virology (LV) located at the Rocky Mountain Laboratories in Hamilton, Montana. LV will interact with four other Intramural Research Laboratories at this location presently studying infectious diseases involving viruses, bacteria, rickettsia, chlamydia and prions.

The mission of the LV is to study high containment BSL-3 and BSL-4 viral pathogens with the goal of developing diagnostics, vaccines, and therapeutics. The research to be conducted in the LV is to include studies of vector/reservoir transmission, pathogenesis, pathophysiology and host immune response of high containment viral pathogens. In addition, the LV must maintain a flexible infrastructure to permit rapid analysis of newly emerging high containment viral pathogens of special interest.

The selected candidate will supervise research in a newly constructed Integrated Research Facility which houses three BSL-4 lab suites, three BSL-3 lab suites and multiple BSL-2 lab suites, as well as extensive associated BSL-2, 3, and 4 animal facilities.

This position requires a Ph.D., M.D., D.V.M. or equivalent with proven ability to carry out a strong independent research program. Preference will be given to candidates with a record of leadership and accomplishment in BSL-4 or Select Agent BSL-3 viral pathogen research, with program(s) consistent with the mission of the NIAID. The selected person will also be expected to recruit and supervise other Principal Investigators with independent research programs.

The Laboratory Chief will have independent resources to conduct laboratory research and translational/clinical research, as appropriate. Committed resources include space, support personnel and an allocated annual budget to cover service, supplies and salaries. A Laboratory Chief in the DIR is equivalent to a Department Chair in a University or Medical School. Applicants must be eligible for the appropriate security clearance under the CDC Select Agent Program. Salary is dependent on experience and qualifications. Interested candidates may contact **Dr. Karyl Barron, Deputy Director, DIR, NIAID at 301/402-2208 or email (kbarron@niaid.nih.gov)** for additional information about the position.

To apply for the position, candidates must submit a curriculum vitae, bibliography, a detailed statement of research interests, the names of three references, and reprints of three selected publications, preferably via email to: **Felicia Braunstein at braunsteinf@niaid.nih.gov or by US Mail to: Ms. Felicia Braunstein, DIR Committee Manager, 10 Center Drive MSC 1349, Building 10, Rm. 4A-30, Bethesda, Maryland 20892-1349**. Please note search **#006** when sending materials. Completed applications **MUST** be received by **Friday, November 3, 2006**. Further information on working at NIAID is available on our website at: **http://healthresearch.niaid.nih.gov**

Chief, Laboratory of Bacterial Diseases
National Institute of Allergy and Infectious Diseases
National Institutes of Health

The National Institute of Allergy & Infectious Diseases (NIAID), Division of Intramural Research (DIR) is seeking an outstanding individual to head the newly established Laboratory of Bacterial Diseases (LBD) in Bethesda, Maryland. The laboratory is to be located in the new C.W. Bill Young Center for Biodefense and Emerging Pathogens located on the NIH campus in Bethesda, Maryland.

The mission of the LBD will be to study basic and applied aspects of bacterial diseases related to biodefense or emerging and re-emerging pathogens, focusing on pathogenic bacteria. Exceptional scientists with research interests in basic, translational or clinical aspects of bacterial pathogenesis are encouraged to apply. The long-term goals of the Institute include supporting research that enables the development of new diagnostics, vaccines, and therapeutics.

This position requires an M.D., Ph.D. or equivalent with proven leadership abilities and a strong independent research program. Preference will be given to candidates with a documented record of accomplishment in bacterial disease research, and those whose program(s) are consistent with the mission of the NIAID.

The Laboratory Chief will have independent resources to conduct basic and clinical research and will supervise other Principal Investigators with independent research programs. The successful candidate is expected to lead a strong research program in laboratory and/or clinical research. Committed resources include space, support personnel and an allocated annual budget to cover service, supplies, animals and related resources and salaries. A Laboratory Chief in the DIR is equivalent to a Department Chair in a University or Medical School. Applicants must be U.S. citizens or permanent residents and be eligible for the appropriate security clearance under the CDC Select Agent Program. Salary will be commensurate with experience and qualifications.

Interested candidates may contact **Dr. Karyl Barron, Deputy Director, DIR, NIAID at 301/402-2208 or email (kbarron@niaid.nih.gov)** for additional information about the position and/or infectious diseases research at the NIH.

To apply for the position, candidates must submit curriculum vitae, bibliography, a detailed statement of research interests, and reprints of up to three selected publications, preferably via Email to: Lynn Novelli at novelly@niaid.nih.gov. In addition, the names of three potential references must be sent to **Dr. Steven M. Holland, Chair, NIAID Search Committee, c/o Ms. Lynn Novelli, DIR Committee Manager, 10 Center Drive, MSC 1356, Building 10, Room 4A26, Bethesda, Maryland 20892-1356.** Completed applications **MUST** be received by **Monday, September 25, 2006**. Please refer to **AD#004** on all correspondence. Further information on this position and guidance on submitting your application is available on our website at: **http://healthresearch.niaid.nih.gov**

WWW.NIH.GOV

NATIONAL INSTITUTE OF MENTAL HEALTH
INTERVENTIONS POSITION AVAILABLE

Are you interested in an exciting, meaningful, and challenging career working with some of the most outstanding scientists in the world? Then the National Institute of Mental Health (NIMH), a major research component of the National Institutes of Health (NIH) and the Department of Health and Human Services, invites you to apply for the position of Health Scientist Administrator in the Division of Extramural Activities, Extramural Review Branch (ERB). This position requires expertise in at least one of the following mental health research areas: HIV AIDS prevention, treatment or preventive interventions, mental health services, psychopharmacology, behavioral medicine, or neuroscience.

The NIMH mission is to reduce the burden of mental illness and behavioral disorders through research on mind, brain, and behavior. The ERB is responsible for the review of the scientific merit of grant applications (e.g., research and training grants, fellowship applications, cooperative agreement concepts and applications), contracts and for concept review of research and development contracts. The successful candidate will join a highly interactive and diverse group of scientists and will be responsible for all aspects of planning, coordinating, directing, and implementing peer reviews of all applications and proposals focusing on one or more of the areas listed in the mandatory selection criteria (see below). You will have the opportunity to meet and work with top scientists in the country and participate in selecting state-of-the-art research in mental health.

Selection Criteria: Experience in clinical research pertaining to prevention research in the area of HIV AIDS, and/or pertaining to the causes, diagnosis, and treatment of mental illness affecting all ages and socio-cultural groups (e.g., etiology, epidemiology, assessment, development/efficacy/effectiveness of psychosocial and/or pharmacologic interventions), and/or experience in the field of neuroscience.

In order to qualify for this career position you should have a Ph.D. and/or M.D. degree in a relevant field of biomedical behavioral science and appropriate, clinical experience. Salary will be commensurate with experience and expertise.

If you are interested in the position described above, please send a letter describing your interest as well as your curriculum vitae to **Henry Haigler, Ph.D., NIMH, NIH c/o Ms. Amita Patel, 6001 Executive Blvd, Room 6-166, Bethesda, MD 20892-9609 or e-mail: apatel@mail.nih.gov**.

Health Scientist Administrator, GS-601-13/14/15
Medical Officer, GS-602-13/14/15
Extramural Program

The National Institute of Arthritis and Musculoskeletal and Skin Diseases (NIAMS) is seeking experienced and highly motivated people to work with the Extramural Program (EP) staff to plan and direct the Institute's research programs in the fields of arthritis, muscle biology, bone biology and bone diseases, musculoskeletal diseases, and skin diseases and to provide independent leadership in research of National and international significance. As a Health Science Administrator in the EP at the NIAMS, the selectee will join a team of professionals responsible for advancing basic, translational and clinical research, research training, and information programs on many of the more debilitating diseases affecting the American people. Areas of research of interest to the EP include autoimmune and inflammatory systemic rheumatic and skin diseases, biology and molecular pathogenesis of connective tissues and related inheritable diseases, degenerative joint disease, orthopedics, bioengineering and biomechanics, basic bone biology and bone diseases; skin biology and pathophysiology; muscle biology and muscle diseases. Candidates with specific experience in one or more of the above listed scientific areas will compete well for these positions.

There are multiple Health Scientist Administrator and Medical Officer vacancies open within the Extramural Program in Bethesda, Maryland. Benefits of the positions include: working in a dynamic research organization and challenging environment; significant involvement in long-range scientific planning and priority-setting; and on-going opportunities for professional development through seminars, formal coursework, and attendance at National meetings.

Salary is commensurate with qualifications and professional experience, and it includes a full Federal benefits package (which includes retirement, health, life and long-term care insurance, Thrift Savings Plan participation, etc.). Physicians may also be eligible to receive a Physicians Comparability Allowance up to $30k per year, depending on qualifications/experience.

For qualifications required, evaluation criteria, and application instructions, view the vacancy announcements at: **http://www.jobs.nih.gov/sciencejobs.asp. Announcement Number: NIAMS-06-139075 for the GS-13/14 Health Scientist Administrator and NIAMS-06-144306 for the GS-15 level Health Scientist Administrator.** Announcement Number: **NIAMS-06-145751 for the GS-13/14 Medical Officer and NIAMS-06-145533 for the GS-15 level Medical Officer.** For additional information on application procedures, call **Lauren Carroll Tedesco at 301-594-2288.** Applications must be received by **October 31, 2006**.

National Institute of Arthritis and Musculoskeletal and Skin Diseases website: http://www.niams.nih.gov/

Department of Health and Human Services
National Institutes of Health
National Institute of Dental and Craniofacial Research
Health Scientist Administrator

We are seeking outstanding scientists, with doctorate level training, independent research, and administrative experience, to join a team of interactive and diverse group of scientists to help shape the future of state-of-the-art research in oral, dental, and craniofacial biology. The National Institute of Dental and Craniofacial Research (NIDCR), a major research component of the National Institutes of Health (NIH) and the Department of Health and Human Services, invites you to apply for the position of Health Scientist Administrator in the Division of Extramural Activities, Scientific Review Branch. The mission of the NIDCR is to improve oral, dental and craniofacial health through research and research training in basic and clinical research on the full spectrum of topics related to oral, dental and craniofacial biology, including oral cancer, chronic pain, immunology, salivary gland physiology, mineralized tissue, craniofacial development and genetics, biomimetics, tissue engineering, and health promotion and behavior.

The incumbent will serve as a Scientific Review Administrator (SRA), and be responsible for the initial administrative and scientific merit review of applications for research programs and/or research training and career development grants through interaction with established scientists in a variety of fields. SRA's are responsible for assuring the fairness and consistency of the scientific peer review process, and for providing technical guidance on peer review policies and procedures and review criteria to applicants, reviewers, and Institute staff.

The salary range for this position is $65,048 to $118,828 per annum, commensurate with qualifications and professional experience. A full benefits package is available, which includes retirement, Thrift Savings Plan participation, health, life and long-term care insurance.

For qualification requirements, evaluation criteria, and application instructions, view the vacancy announcements at **http://jobsearch.usajobs.opm.gov/a9nih.asp**. Refer to announcement **# NIDCR-06-145299DE** and **NIDCR-06-148542MP**. Applications will be accepted until **October 20, 2006**. Please contact **Michelle Lipinski at 301-594-2286 or lipinskim@od.nih.gov** if you have questions.

Department of Health & Human Services (DHHS)
National Institutes of Health (NIH)
National Institute of Dental and Craniofacial Research (NIDCR)

The National Institute of Dental and Craniofacial Research (NIDCR), National Institutes of Health (NIH), Department of Health & Human Services (DHHS) is seeking applicants for a Biologist/Microbiologist/Health Scientist Administrator position in the Center for Integrative Biology and Infectious Diseases (CIBID). The position advertised is for the Director of the Microbiology Program. This program supports extramural basic and translational research on the role of oral microbes in health and disease. To this end, four broad scientific areas provide the basis for rapid development of knowledge of the etiology, pathogenesis, diagnosis, treatment and prevention of oral infectious diseases. These interrelated areas are: (i) Biofilms and Microbial Ecology; (ii) Microbial genomics; (iii) Microbial Virulence and Disease Pathogenesis; and (iv) Prevention and Treatment.

The incumbent will direct, administer and evaluate a portfolio of extramural grants, contracts and cooperative agreements and will stimulate interest in and provide advice to the extramural community regarding the respective research portfolio. In addition, the incumbent will participate in funding decisions, policy development, as well as implementation and coordination with other programs both within and outside of the NIDCR.

The salary range for this position is $77,353 to $118,828 per annum, commensurate with qualifications and professional experience. A full benefits package is available, which includes retirement, Thrift Savings Plan participation, health, life and long-term care insurance.

For qualifications required, evaluation criteria, and application instructions, view the vacancy announcements at: **http://jobsearch.usajobs.opm.gov/a9nih.asp**. Refer to announcement **# NIDCR-06-141634DE** or **NIDCR-06-147841MP**. Applications will be accepted until **October 27, 2006**. Please contact **Michelle Lipinski at 301-594-2286** or **lipinskim@od.nih.gov** if you have questions.

WWW.NIH.GOV

SCIENTIFIC PROGRAM ANALYST POSITION
NIH Roadmap Molecular Libraries and Imaging Initiative

A Scientific Program Analyst position is available for the Roadmap Molecular Libraries and Imaging Initiative, an NIH effort aimed at building a national capability to develop pharmacological probes for investigating biological function **(see: http://nihroadmap.nih.gov/)**. The position will provide support to the Program Director for the Initiatives' *Assay Development for High Throughput Screening* Program. This Program is rapidly developing a large and diverse portfolio of assay projects aimed at pharmacological probe development, and it will be a key role of this position to collect, analyze and evaluate data allowing the cross-Institutes impact of these funded projects to be visualized. This person will further assist in the tracking and analysis of applications and funded grants, contact with investigators including response to inquiries and requests for information, development of funding plans and implementation of administrative supplements, and the preparation and collection of Program evaluation criteria.

The ideal candidate will have an advanced degree in the Biological Sciences and postdoctoral experience. Experience with the techniques of modern molecular, biochemical, and/or cell biology research is a must. Knowledge of cellular pharmacology and drug screening would be highly desirable. Familiarity with Microsoft Word, Excel and Powerpoint will be expected. For additional information about the position please contact Program Director **Mark Scheideler, Ph.D.** by email at: **scheidelerm@ninds.nih.gov** . This position is available from **October 1, 2006**.

Tenured/Tenure-Track Position
Neuroimmunology (Clinical)
Division of Intramural Research

The Division of Intramural Research of the National Institute of Neurological Disorders and Stroke is recruiting an individual for a tenured or tenure-track position in the area of neuroimmunology with a focus on clinical research. The applicant should have a special interest and experience in translational clinical research relating to multiple sclerosis or other immune mediated disease of the central nervous system. The individual would direct an independent research program on immune mediated diseases of the nervous system and especially multiple sclerosis. The program would conduct its work in conjunction with the Neuroimmunology Branch (NIB) which was established to study the cause and treatment of immunological mediated diseases of the central nervous system. The individual should have a demonstrated background and knowledge in research focused on immune mediated disease of the nervous system and with expertise in human immunology and/or the application of clinical trial methodology to the study of disease mechanisms and testing new therapies. The candidate will have earned a M.D. degree and will have excellent scientific skills in structuring an original and productive research program using outstanding communication and collaborative abilities. The candidate will have a medical license in the United States and preference will be given to a candidate who has completed training in an accredited training program in neurology and is either board eligible or board certified in Neurology. Candidates for a tenured position must have an international reputation and well-documented evidence of ongoing independent accomplishments. An individual selected for a tenure-track position is expected to build a dynamic and productive research group. Laboratory facilities, start-up and sustained research funds and salary will be competitive with premier academic institutions. Applicants should send curriculum vitae, bibliography, statement of research interests, and names of references to: **Dr. Story Landis, Director, National Institute of Neurological Disorders and Stroke, c/o Peggy Rollins, Office of the Scientific Director, Division of Intramural Research, Building 35 Room GA908, NIH, Bethesda, MD 20892**. Applications will be reviewed upon receipt.

Tenured/Tenure-Track Position
Neuroimmunology (Basic/Translational)
Division of Intramural Research

The Division of Intramural Research of the National Institute of Neurological Disorders and Stroke is recruiting an individual for a tenured or tenure-track position in the area of Neuroimmunology. The applicant should have a special interest and experience in translational research relating to multiple sclerosis or other immune mediated disease of the central nervous system. The individual would direct an independent research program on molecular, biological or immunological aspects of immune mediated diseases of the nervous system and especially multiple sclerosis. The program would conduct its work in conjunction with the Neuroimmunology Branch (NIB) which was established to study the cause and treatment of immunological mediated diseases of the central nervous system. The individual should have a demonstrated background and knowledge in research focused on immune mediated disease of the nervous system and with expertise in the use of animal models or in human immunology. The candidate will have a Ph.D. and/or M.D. degree with excellent scientific skills in structuring an original and productive research program using outstanding communication and collaborative abilities. Candidates for a tenured position must have an international reputation and well-documented evidence of ongoing independent accomplishments. An individual selected for a tenure-track position is expected to build a dynamic and productive research group. Laboratory facilities, start-up and sustained research funds and salary will be competitive with premier academic institutions. Applicants should send curriculum vitae, bibliography, statement of research interests, and names of references to: **Dr. Story Landis, Director, National Institute of Neurological Disorders and Stroke, c/o Peggy Rollins, Office of the Scientific Director, Division of Intramural Research, Building 35 Room GA908, NIH, Bethesda, MD 20892 (301-435-2232)**. Applications will be reviewed upon receipt.

Staff Scientist

Section of Molecular Mechanisms of Glaucoma, Laboratory of Molecular and Developmental Biology, National Eye Institute, National Institutes of Health, Department of Health and Human Services is seeking applications for a Staff Scientist with experience and established track record in molecular, cellular and developmental biology, with particular emphasis on neuroscience, vision research, and protein chemistry. Research projects include rodent models of glaucoma, identification and characterization of genes involved in glaucoma, and elucidation of molecular changes in eye tissues with glaucoma progression. The position is a term appointment, renewable indefinitely upon mutual agreement, and is intended to be long term, with the individual playing a major role in maintaining research continuity within the laboratory. This is not a principal investigator position and is not endowed with independent funding or space. Salary is commensurate with the degree of relevant experience and training in the field, with the range of $73,178-107,403 per annum. Applicants must have a Ph.D. or M.D. degree. Interested candidates should send a statement of research interests and goals, curriculum vitae, bibliography, and contact information for three references to **Dr. Stanislav Tomarev, Laboratory of Molecular and Developmental Biology, National Eye Institute, NIH, Building 7, Room 103, 7 Memorial Drive, MSC 0704, Bethesda, Maryland 20892-0704. Tel. 301-496-8524; Fax 301-496-8760; e-mail: tomarevs@nei.nih.gov**.

UCDAVIS
School of Veterinary Medicine

Assistant/Associate Professor of Veterinary Anatomic Pathology (tenure track)

Veterinarian with advanced training in anatomic pathology required. Eligibility for certification by the American College of Veterinary Pathology required. Board certification preferred. PhD required at time of appointment. Clinical experience and competence in anatomic pathology. Demonstrated aptitude/experience in teaching. Documented research record or potential to develop an independent research program using contemporary technologies for the characterization of pathogenesis of disease at a cellular and/or molecular level is essential. Must possess excellent interpersonal and communication skills and a demonstrated ability to work with others in a collegial team atmosphere.

To receive fullest consideration, applications must be received by **November 1, 2006**; position open until filled. Expanded position description at **http://www.vetmed.ucdavis.edu/pmi/PMIpage1. htm**. Submit letter of intent outlining special interest in the position, overall qualifications, experience, and career goals; CV; and names and addresses of three professional references to: **Dennis W. Wilson, Chairman, Department of Pathology, Microbiology and Immunology, School of Veterinary Medicine, University of California, Davis, Davis, CA 95616, Attn: Donna Roggenkamp**.

AA/EOE

Assistant Professor Position Biochemistry and Structural Biology Department of Molecular and Cellular Biology, Harvard University

As part of a broad expansion in the Life Sciences at Harvard University, the Department of Molecular and Cellular Biology has an opening for the tenure-track position of Assistant Professor in the fields of **biochemistry and structural biology**. Our department covers a broad range of topics, including molecular biology, cellular biology, developmental biology, neurobiology, molecular evolution, systems biology, biochemistry, and structural biology. The department provides access to many core facilities, including imaging, proteomics, genomics, and bioinformatics. Harvard University is part of the NE-CAT consortium at Argonne National Laboratories.

Applications should include: curriculum vitae, reprints of publications, and a statement of present and future research plans (1-3 pages). Complete applications and three letters of recommendation, solicited by the applicant, should be received not later than **10 November 2006**. Submit applications to **BSB@mcb.harvard.edu** or to: **J. Blackbourn/MCB Search Committee, Department of Molecular and Cellular Biology, Harvard University, 7 Divinity Avenue, Rm. 167, Cambridge MA 02138.**

Harvard is an Affirmative Action/Equal Opportunity Employer. We strongly encourage applications from women and minority candidates.

www.mcb.harvard.edu

PURDUE UNIVERSITY

Faculty Positions in Biology, Chemistry, Computer Sciences, Earth & Atmospheric Sciences, Mathematics, Statistics, and Physics with possible joint appointments in Agriculture, Education, and Engineering

Purdue University: Where the Sciences Coalesce

COALESCE FACULTY POSITIONS

Purdue University's College of Science, as part of a University-wide initiative to target compelling national research priorities that require insights and contributions from multiple disciplines, is adding 60 additional new multidisciplinary faculty positions during this decade. We are seeking applicants with research and teaching excellence for each of the seven areas of research coalescence identified in the College's strategic plan.

- **Bioinformatics:** systems biology, computer science, comparative and/or agricultural genomics, modeling of biological data, computational methods.
- **Climate Change:** searches in ecohydrology (joint with Agriculture and Engineering) and Aerosols, Clouds, and Radiation (joint with Agriculture).
- **Computational Science:** searches in applied mathematics, high performance computing and advanced computational methods.
- **Massive Data:** searches in visualization and spatial statistics (joint with Agriculture).
- **Membrane Science:** searches in biochemistry and structural biology of membrane proteins, vesicle trafficking, and biophysics of membranes.
- **Nanoscience:** searches in transport in nanostructures, advanced imaging at the nanometer scale, computational nanoscience.
- **Science Education Research:** searches in education research in Biology, Computer Science, Mathematics, or Physics, although outstanding applicants in other disciplinary areas will be considered.

All hires will have a departmental home, but hires may be joint between departments or other colleges.

CORE FACULTY POSITIONS

Department of Biological Sciences
Faculty position in Cancer Cell Biology
Faculty position in Vertebrate Developmental Biology

Department of Chemistry-Biochemistry
Faculty position in Biochemistry

Department of Computer Science
Faculty position in Networking

Department of Earth and Atmospheric Sciences
Faculty position in Geodynamics and Active Tectonics

Department of Mathematics
Faculty position in Pure Mathematics

Department of Physics
Faculty position in Experimental High Energy Nuclear Physics
Faculty position in Theoretical Biological Physics

Department of Statistics
Faculty position in High Dimensional Theoretical Statistics

A Ph.D. in a field related to the position sought is required. The successful candidates will teach courses and conduct research in their area of specialty.

For more information about the Purdue University College of Science, its areas of coalescence, and how to apply for a faculty position, visit our Web site at **http://www.science.purdue.edu/FACULTYSEARCHES/**. Information about related searches in other departments will also be posted there.

Purdue University is an Equal Opportunity/Affirmative Action Employer fully committed to achieving a diverse work force.

ASSISTANT PROFESSOR, BIOLOGY
LOYOLA COLLEGE IN MARYLAND

The Biology Department at Loyola College is offering a tenure track Assistant Professor position to begin in the fall of 2007. Applicants should be trained as a microbiologist with expertise in at least one of the following areas: molecular biology, medical microbiology, or virology. The successful candidate will be given the opportunity to teach introductory and upper-level courses in biology, be engaged in research involving undergraduate students, participate in departmental and college-wide service, and to receive guidance and mentoring from members of the department. The successful applicant will be expected to teach introductory courses in Cell and Molecular Biology, Organismal Biology, and/or Ecology, Evolution and Diversity, and also to teach upper level courses in their areas of specialization. Preference will be given to candidates with a strong background in cell/molecular biology or virology. Candidates should have a Ph.D in Biology or a related discipline. Post-doctoral training and college teaching experience are preferred.

For more information about Loyola, please review our websites at:
http://www.loyola.edu/ and **http://www.loyola.edu./biology/**

Please apply electronically at **www.careers.loyola.edu** and include a curriculum vitae, statement of teaching and research interests, a teaching philosophy, and contact information for three references. For more information please contact **Monika Matthews** (**mmatthews@loyola.edu**), Administrative Assistant to the Biology Department or **Dr. Kim Derrickson** (**kderrickson@loyola.edu**), Search Committee Chair of the Biology Department. Applications received by **October 15, 2006** will receive full consideration.

Loyola College is a selective liberal art, Jesuit Catholic institution that welcomes applicants from all backgrounds who can contribute to its educational mission. Loyola College is an Equal Opportunity Employer, seeking applicants from underrepresented groups.

Assistant or Associate Professor
BIOMEDICAL ENGINEERING

The Department of Biological Engineering at the University of Missouri – Columbia invites applications for a tenure-track or tenured faculty position. Candidates with research strengths related to bioMEMS/NEMS and microfluidics applied to problems of cell physiology are preferred, however other areas that complement existing strengths in the department will be considered. The successful candidate will also teach at the undergraduate and graduate levels. Competitive salary, start-up package, and laboratory facilities will be provided.

MU offers a rich environment for collaboration and Columbia is consistently ranked as one of the top 20 places to live in the U.S. The MU Department of Biological Engineering is rapidly expanding with faculty expertise in biosensors, bioMEMS, biomaterials, electrophysiology, biomechanics, and biophotonics.

Applicants should have an earned doctoral degree in biomedical engineering or a related field and a strong background in both engineering and life sciences. Senior-level candidates are expected to have a vigorous, extramurally funded research program, whereas candidates applying at the Assistant Professor level must have strong potential for establishing an externally funded research program. Postdoctoral training is required except under special circumstances. Review of applications will begin immediately and will continue until the position is filled.

Applicants should submit Curriculum Vitae, summary of past research and future research plans, brief statement of teaching plans, and list of 3-5 professional references to: **Search Committee Chair, Department of Biological Engineering, 215 Agricultural Engineering Building, University of Missouri-Columbia, Columbia, MO 65211. Ph: (573) 882-2369; Email: RatliffDe@missouri.edu**.

MU is an Equal Opportunity-Affirmative Action Employer.

Penn
University of Pennsylvania

The Department of Dermatology at the University of Pennsylvania's School of Medicine seeks candidates for an Assistant, Associate and/or Full Professor position in the tenure track. Rank will be commensurate with experience. Responsibilities include some patient care, as well as resident, fellow, and medical student education. Applicants must have an M.D. or M.D./Ph.D. degree and have demonstrated excellent qualifications in clinical care, education, and research. Board Certified in Dermatology.

Current research interests in the Department include: differentiation, adhesion, embryological development, stem cells and signal transduction in epidermis and hair follicles; gene therapy targeting the epidermis and hair follicle; microRNA function in the skin; basic studies of autoimmune blistering and rheumatologic diseases of skin, impetigo and staphylococcal scalded skin syndrome; proteases in skin physiology and pathophysiology; and basic pathophysiologic and immunologic studies of cutaneous T cell lymphoma.

Please submit curriculum vitae, a brief statement of research interest, and three reference names to:

Sarah E. Millar, Ph.D.
Chair, Search Committee
Department of Dermatology
University of Pennsylvania
School of Medicine
422 Curie Boulevard, M8D SCL
Philadelphia, PA 19104
Email: millars@mail.med.upenn.edu
Fax: (215) 573-2033

The University of Pennsylvania is an Equal Opportunity, Affirmative Action Employer. Women and minority candidates are strongly encouraged to apply.

ASSISTANT PROFESSOR
ENVIRONMENTAL HEALTH

The Department of Earth and Environmental Science in the College of Science at the University of Texas at Arlington invites applications for a tenured/tenure-track faculty position. This position is at the rank of tenure-track Assistant Professor. Candidates with outstanding credentials will be considered for appointment as tenured Associate or Full Professor. This position has a starting date of September 1, 2007.

An earned doctorate in a discipline relating to Environmental Health is required by the starting date, and preference will be given to candidates with research experience in an area of Toxicology and demonstrated capability to conduct interdisciplinary research and obtain extramural funding. The position comes with high quality research laboratory space and a substantial startup package. The successful candidate will initiate a vigorous extramurally funded research program, supervise graduate students, and teach graduate and undergraduate courses in areas relating to Environmental Health.

The University of Texas at Arlington is located in the center of the Dallas-Fort Worth metropolitan area. The department (**http://www.uta.edu/geology/**) is home to a rapidly expanding interdisciplinary graduate program in Environmental and Earth Sciences offering the M.S. and Ph.D. (**http://www.uta.edu/ese/**). Applications should consist of a curriculum vitae (listing education, positions held, publications, grants and contracts, etc.) and statements outlining research and teaching interests. Applications will be accepted only by e-mail as a pdf attachment sent to **envhealth@uta.edu**. A list of at least three professional references should be provided. The search committee will begin reviewing applications immediately, and the position will remain open until filled. Final offer of employment is contingent on completion of a satisfactory criminal background investigation for security sensitive positions.

UT Arlington is an Equal Opportunity/Affirmative Action Employer.

MOLECULAR PATHOLOGY LABORATORY DIRECTOR - PVAMC

Penn
University of Pennsylvania

The Department of Pathology and Laboratory Medicine at the University of Pennsylvania's School (UPenn) School of Medicine and the Philadelphia Veterans Administration Medical Center (PVAMC) seek candidates for an Assistant, Associate and/or Full Professor position in either the tenure track or the non-tenure clinician-educator track. Rank and track will be commensurate with experience. The successful applicant will have experience in the field of Molecular Pathology. Applicants must have an M.D. or M.D./Ph.D. or equivalent degree and have demonstrated excellent qualifications in education, research, and clinical care. Fellowship training with board certification/eligibility in Molecular Genetic Pathology (MGP, ABP) or Clinical Molecular Genetics (ABMG), or equivalent training and experience, is required.

Responsibilities include directing a Clinical Molecular Pathology Laboratory and a Molecular Core Facility at the PVAMC and serving as an attending physician at the Molecular Pathology Laboratory at the Hospital of the University of Pennsylvania (HUP) with oversight responsibility of laboratory personnel. Teaching and mentoring of medical students, biomedical graduate students, pathology residents, and fellows, and technologists will be an integral component of the position.

The successful candidate will be expected to develop and direct a new molecular diagnostic lab at the PVAMC which serves as a referral center for the Eastern Market of VA Network 4. A primary responsibility will be to develop and validate clinical molecular tests focusing on infectious diseases, inherited disorders, pharmacogenomics, and molecular oncology diagnosis and classification, with a particular emphasis on sequencing-based technologies. An additional responsibility will be to assist researchers at the PVAMC in the use of the core molecular facility. We seek a dynamic person with experience in Clinical Molecular Pathology, DNA sequencing, and an interest in the development and implementation of molecular diagnostic clinical assays. The candidate must demonstrate a strong record in clinical, translational or basic research and will be expected to pursue active research and participate in collaborative ventures at the UPenn and PVAMC. C-E track candidates must demonstrate a strong record/potential in clinical, translational or collaborative research. Tenure track candidates must demonstrate a strong record/potential to garner extramural funding for basic research.

Please submit curriculum vitae, a brief statement of research interests, and three reference letters to:
Paul H. Edelstein, M.D., Director of Clinical Microbiology, Hospital of the University of Pennsylvania
and Pratap M. Yagnik, M.D., PVAMC
c/o Robyn Richardson-Bey, Philadelphia VA Medical Center, Human Resources (05)
3900 Woodland Avenue, Philadelphia, PA 19104
PVAMCJobs@va.gov – Ref: 230-06

The University of Pennsylvania and PVAMC are Equal Opportunity, Affirmative Action Employers.
Women and minority candidates are strongly encouraged to apply.

St. Jude Children's Research Hospital
ALSAC • Danny Thomas, Founder
Finding cures. Saving children.

St. Jude Children's Research Hospital (SJCRH), a premier center for biomedical investigation located in Memphis, Tennessee, USA, is seeking to fill three positions in the Department of Structural Biology. Research in the Department is highly interactive within the hospital and centers on understanding the molecular basis of biological processes and human diseases. Examples of ongoing research include mechanistic studies of protein degradation, cell cycle regulation, tumor suppressor function, signal transduction, transcriptional regulation, DNA repair, structure-based drug design and lipid metabolism. A wide range of structural techniques are available in-house, including X-ray crystallography, NMR spectroscopy and high-level computing. Regular access to synchrotron radiation is guaranteed through membership of SER-CAT at the Advanced Photon Source.

SJCRH is a hospital and basic research institute that focuses on the fundamental causes and treatment of catastrophic childhood diseases including cancer, infectious diseases and genetic disorders. Founded by Danny Thomas in 1962, the hospital currently includes some 150 basic and clinical investigators organized into a traditional academic environment. The research environment at SJCRH is highly interactive, with opportunities to collaborate with investigators in other Departments, including Biochemistry, Chemical Biology and Therapeutics, Developmental Neurobiology, Genetics and Tumor Cell Biology, Hematology, Oncology, Immunology, Infectious Diseases, Molecular Pharmacology, Pathology and Pharmaceutical Sciences. All investigators have access to state-of-the-art core facilities that include proteomics, genomics, bioinformatics, imaging, protein production, molecular synthesis and high-throughput small molecule screening. SJCRH continues to receive support through the fundraising efforts of the American Lebanese Syrian Associated Charities (ALSAC).

Position 1 - Assistant Member
The candidate will be expected to develop an independent and funded research program and to eventually become an established investigator within the hospital. He/she will have an interest in applying structural/biophysical methods to address fundamentally important biological questions. This new position will be supported by generous startup funds and personnel.

Position 2 - Staff Macromolecular Crystallographer
The candidate will have expertise in all aspects of crystallography with the primary duty of supporting research in the department. Expertise in the use and implementation of state-of-the-art crystallographic software is essential, and extensive experience with de novo structure determination is required. Familiarity with high-throughput crystallization apparatus would be useful. The staff member will also be expected to accompany and assist members of the department with synchrotron data collection.

Position 3 - Staff NMR Spectroscopist
The candidate will have expertise in all aspects of state-of-the-art NMR spectroscopy with the primary duty of supporting research in the department. In particular, we seek an individual interested in implementing new solution protein NMR techniques, and to be involved in evaluating, procuring and utilizing new NMR hardware/software, e.g. in areas such as solid-state NMR of proteins and NMR analysis of the human metabolome.

Candidates for the two staff positions should have excellent communication skills and will be involved in the training of junior members of the department in all aspects of crystallographic and NMR structure determination. There will also be opportunities for collaborative studies with departmental faculty members.

Candidates for all three positions should have a PhD and/or MD degree, at least three years of relevant postgraduate experience, and a demonstrated track record of productivity. **Applicants should send a curriculum vitae, a 1-2 page summary of research interests and future plans, and the names of three references to: Dr. Stephen W. White, Chair, Department of Structural Biology, St. Jude Children's Research Hospital, 332 N. Lauderdale, Memphis, TN 38105.**

www.stjude.org

St. Jude is an Equal Opportunity Employer and a Drug-Free Workplace.
Candidates receiving offers of employment will be subject to preemployment drug testing and background checks.

FACULTY POSITION
IN
Molecular Biotechnology
UNIVERSITY OF COLORADO, BOULDER

The University of Colorado invites applications for tenure-track faculty positions in the broad area of molecular biotechnology, under the auspices of the Initiative in Molecular Biotechnology, a program that integrates faculty from the departments of Applied Mathematics, Chemical and Biological Engineering, Chemistry and Biochemistry, Computer Science, Molecular, Cellular and Developmental Biology, and Physics (**http://bayes.colorado.edu/biotech**). Individuals with interests in biological systems are encouraged to apply. Specific areas of interest include but are not limited to bioanalytical, biosensor design, cell imaging, chemical biology, RNA biology, tissue and stem cell engineering. Candidates at the **ASSISTANT, ASSOCIATE**, and **FULL PROFESSOR** levels will be considered. Candidates must have a Ph.D. degree and enthusiasm for teaching at undergraduate and graduate levels, and will be expected to develop an internationally recognized research program.

Applicants should submit a curriculum vitae, statements of research and teaching interests, and arrange to have three letters of reference sent to **Biotechnology Search Committee Chair, Campus Box 347, University of Colorado, Boulder, CO 80309-0347**. Electronic submission of all application materials may be sent to **BioTApp@colorado.edu** Applicants should indicate their preferred home department. For full consideration, applications should be received by **November 15, 2006**, although all applications will be reviewed until the positions are filled.

The University of Colorado is sensitive to the needs of dual career couples. The University of Colorado at Boulder is committed to diversity and equality in education and employment.

SLOAN-KETTERING INSTITUTE
CELL BIOLOGY PROGRAM
Tenure-Track Faculty Positions

The Cell Biology Program, Sloan-Kettering Institute (www.ski.edu) has initiated a search for tenure-track faculty members. We are interested in outstanding individuals who have the potential to develop an innovative, independent research program that complements and enhances our existing strengths. Candidates with research interests in exciting areas of eukaryotic cell biology and using a variety of experimental approaches and systems are encouraged to apply. New faculty will be eligible for appointment in the recently established Gerstner Sloan-Kettering Graduate School of Biomedical Sciences as well as the Weill Graduate School of Medical Sciences of Cornell University. Sloan-Kettering has an outstanding infrastructure and state of the art core resources. The new 23-story Zuckerman Research Center opened earlier this year will allow significant expansion of our research programs.

Interested individuals should e-mail their application, preferably in PDF format, to **cellbio@mskcc.org** by November 17, 2006. The application should include a CV, a description of past research, a description of proposed research, and copies of three representative publications. Candidates should arrange to have three signed letters of reference sent by e-mail to **cellbio@mskcc.org** or by mail to: **Stephanie Miranda, Coordinator, Cell Biology Program, 1275 York Avenue, Box 428, New York, NY 10021.** Specific inquiries can also be made by e-mail to **cellbio@mskcc.org.** EOE/AA.

Memorial Sloan-Kettering Cancer Center
The Best Cancer Care. Anywhere.
www.mskcc.org

JOHNS HOPKINS
MEDICINE
SCHOOL OF MEDICINE

Faculty Positions
The Solomon H. Snyder Department of Neuroscience

Applications are invited for tenure-track faculty positions at both junior and senior levels in the Solomon H. Snyder Department of Neuroscience at the Johns Hopkins University School of Medicine. Applicants should have broad interests in molecular, cellular, developmental, systems or behavioral neuroscience, a Ph.D. or M.D., and a strong record of research accomplishments. Applicants using in vivo imaging and electrophysiological recording approaches, or applicants using zebrafish or invertebrate model organisms, are especially encouraged to apply. Faculty members are expected to have, or establish, active independent research programs and participate in teaching graduate and medical students. Deadline for applications is **November 30, 2006**.

Please submit a PDF file containing curriculum vitae, names and contacts for three references and a brief description of current and future research interests to:

Richard L. Huganir, Ph.D.
Search Committee
Department of Neuroscience
The Johns Hopkins University
School of Medicine
725 North Wolfe Street, PCTB 904
Baltimore, Maryland 21205
JHUNeuroscience@jhmi.edu

An EEO/AA Employer.

ASSISTANT/ASSOCIATE PROFESSOR

Department of Cell & Developmental Biology

Applications are invited for tenure-track positions at the Assistant or Associate Professor level in the Department of Cell & Developmental Biology at SUNY Upstate Medical University in Syracuse. The Department is undergoing a major expansion of its faculty. To complement and enhance the existing research interests, we will be recruiting into the general areas of cellular function and development. At this time we are particularly interested in outstanding candidates with expertise in cardiovascular development and cell signaling, but welcome applications from individuals with research programs in areas of cell motility and the cytoskeleton, cell differentiation, and developmental neurobiology. Substantial renovation and expansion of departmental research space and core facilities has begun in order to support the department's growth.

Candidates should have a Ph.D. and/or M.D. degree, and postdoctoral experience. Assistant Professors will be expected to develop an independent research program, while applicants at the Associate Professor level should have an established track record of research productivity and funding. Substantial startup packages and competitive salaries will be provided to all successful candidates. All faculty will participate in the training and teaching of graduate and medical students.

Please submit CV, descriptions of research accomplishments, future plans for research and teaching interests as a single PDF file to **fontanek@upstate.edu**. Please have three letters of recommendation sent to:

Dr. Christopher Turner, Chair Search Committee
Department of Cell & Developmental Biology
SUNY Upstate Medical University
750 East Adams Street, Syracuse, NY 13210

For additional information, visit the departmental website
www.upstate.edu/cdb

State University of New York
Upstate Medical University
Formerly known as SUNY Health Science Center

An AA/EEO/ADA employer, committed to excellence through diversity.

POSITIONS OPEN

THEORETICAL BIOLOGIST TENURE-TRACK FACULTY POSITION
University of Toronto at Mississauga

The University of Toronto at Mississauga (UTM), Department of Biology, invites applications for a tenure-track faculty position at the level of **ASSISTANT or ASSOCIATE PROFESSOR,** effective July 1, 2007.

The area of specialization is open but we are searching for a Theoretical Biologist who is asking cutting-edge questions in ecology and evolutionary biology. We are particularly interested in cross-disciplinary approaches designed to produce a more broadly integrated conceptual framework.

The successful candidate may use computer modeling, laboratory manipulations, or field-based experiments (or a combination) to answer those questions. Greatest weight, however, will be given to the scope and novelty of the question or questions being asked. Excellent opportunities exist for collaboration both within the Department of Biology, and in other departments with strong research ties to biology. The successful applicant will have a Ph.D., and an outstanding academic record, and evidence of excellence in teaching. Salary will be commensurate with qualifications and experience.

Applicants should provide curriculum vitae, a statement of teaching philosophy and interests, an outline of their proposed research, and should arrange to have three confidential letters of recommendation sent on their behalf to: **Professor Robert Reisz, Chair, Department of Biology, University of Toronto at Mississauga, Mississauga, Ontario, Canada L5L 1C6. E-mail: rreisz@utm.utoronto.ca.** Closing date for submissions is November 15, 2006. For more information on the Department go to **website: http://www.utm.utoronto.ca/~w3bio/homepage/.**

The University of Toronto is strongly committed to diversity within its community and especially welcomes applications from visible minority group members, women, Aboriginal persons, persons with disabilities, members of sexual minority groups, and others who may contribute to the further diversification of ideas.

All qualified candidates are encouraged to apply; however, Canadians and permanent residents will be given priority.

ASSISTANT PROFESSORS OF BIOLOGY
State University of New York at Geneseo

The Department of Biology at State University of New York (SUNY), Geneseo, invites applications for two new tenure-track faculty positions at the rank of Assistant Professor to begin in fall 2007. Candidates should demonstrate potential for excellence in undergraduate teaching and the development of a research program involving undergraduates. Position one: We seek an individual with experience in the ecology of animal or microbial systems, especially those employing molecular or biochemical techniques. Teaching responsibilities include a second-year principles of ecology course and an organismal diversity course. The successful candidate will also contribute to the development and instruction of a laboratory in ecology. Position two: We seek an individual with experience in the cell/molecular mechanisms of physiological and/or developmental processes emphasizing techniques applicable to the study of proteins and working in an animal system. Teaching responsibilities include a course in cell biology lecture and/or laboratory, animal physiology or developmental biology, and possibly advanced laboratories in cell/molecular topics. Online application at **website: http://jobs.geneseo.edu** is required. Please attach a cover letter that includes a list of relevant coursework, curriculum vitae, and statements of teaching philosophy and research interests. In addition, three letters of recommendation and up to three recent reprints should be sent to: **Search Committee, Dr. Ray Spear, Chair, Department of Biology, State University of New York, Geneseo, 1 College Circle, Geneseo, NY 14454.** Review of applications begins October 16, 2006, and continues until the positions are filled. *All applicants are subject to drug and criminal background checks. SUNY Geneseo is an Affirmative Action/Equal Opportunity Employer committed to recruiting, supporting, and fostering a diverse community of outstanding faculty, staff, and students.*

POSITIONS OPEN

Massachusetts Institute of Technology

FACULTY POSITION IN CANCER BIOLOGY

The Center for Cancer Research (CCR) at the Massachusetts Institute of Technology (MIT) is one of eight basic NCI-designated cancer centers. The CCR and Department of Biology at MIT invite applications for a **JUNIOR FACULTY** appointment in the area of experimental cancer biology. Areas of special interest include, but are not limited to: cell migration and metastasis, angiogenesis, stem cell biology, animal models of cancer, cancer pathways, proteomics and intracellular signaling, chemical and systems biology, and cancer therapeutics. The candidate will be expected to lead an innovative research program as well as participate in undergraduate and graduate teaching. Ph.D., M.D., and M.D./Ph.D. candidates are encouraged to apply.

Applicants please submit curriculum vitae, brief summaries of past accomplishments, and a description of future research plans. Letters of recommendation should be sent separately from three scientists who can provide an evaluation of the candidate's accomplishments and future potential for both research and teaching. Please send applications and letters to:

Professor Jacqueline Lees
c/o Lori Spindler
Massachusetts Institute of Technology
E17-110, Center for Cancer Research
77 Massachusetts Avenue
Cambridge, MA 02139-4307

Completed applications will be considered beginning October 1, 2006, and through December 1, 2006.

MIT is an Affirmative Action/Equal Opportunity Employer.

The Department of Chemistry at the University of Michigan invites applications for an anticipated position at the rank of **ASSISTANT PROFESSOR or ASSOCIATE PROFESSOR** in any subdiscipline of chemistry with a proposed start date of September 1, 2007. This would be a University-year appointment (nine months academic salary with three months research supported salary). Candidates are expected to develop an internationally recognized program of scholarly research and to excel in teaching at undergraduate and graduate levels. Detailed information regarding the electronic application process and required materials is available online at **website: http://www.chem.lsa.umich.edu/chem/facultyrecruit/.** The position will remain open until filled but preference will be given to applicants who have submitted all requested materials prior to October 15, 2006. Information about the Chemistry Department is available on the **website:http://www.umich.edu/~michchem.** Questions about the application process should be sent to **e-mail: chemfac06@umich.edu.** *Women and minorities are encouraged to apply. The University of Michigan is supportive of the needs of dual-career couples and is a nondiscriminatory, Affirmative Action Employer.*

The Department of Animal Sciences, Purdue University, West Lafayette, Indiana, invites applications for a **TENURE-TRACK FACULTY POSITION** in adipose biology, available July 2007. Suitable candidate must possess a Ph.D. in animal sciences, cell biology, or a related area; postdoctoral experience is preferred. The successful candidate will be expected to develop an innovative, extramurally funded, and internationally recognized research program on aspects of adipose biology and its role in growth or disease. The candidate will teach and advise both undergraduate and graduate students. Detailed position announcement and application details can be found at **website: http://www.ansc.purdue.edu/positions/AdiposeBiology2006.pdf.** Review of applications will begin November 15, 2006, and will continue until the position is filled. *Purdue University is an Equal Opportunity/Equal Access/Affirmative Action Employer.*

POSITIONS OPEN

The Biology Department of Grand Valley State University invites applicants for two tenure-track faculty positions, at the rank of **ASSISTANT PROFESSOR,** to join an interdisciplinary faculty and rapidly expanding program. Preference will be given to applicants with demonstrated success in teaching and research involving undergraduate or graduate students. Excellent communication skills are required. Complete applications will include letter of application, curriculum vitae, statements of teaching philosophy and research interests, copies of transcripts, and three letters of reference. Deadline for receipt of complete applications is November 20, 2006.

TERRESTRIAL ECOSYSTEM RESTORATION AND MANAGEMENT. Applicants must have a Ph.D. in a natural resources field with expertise in terrestrial ecosystem restoration and management. Additional competence desired in soil science, land reclamation, and related terrestrial ecosystem management applications. Teaching responsibilities include undergraduate courses in soils and land reclamation, a graduate-level course in terrestrial ecosystem restoration, and may include other courses in the candidate's areas of expertise that will enhance our undergraduate and graduate programs. Submit materials to: **Dr. Neil W. MacDonald, Chair, Natural Resources Management Search Committee, Department of Biology, Grand Valley State University, Allendale, MI 49401-9403, telephone: 616-331-2697, e-mail: macdonan@gvsu.edu, website: http://www.gvsu.edu/biology).**

BEHAVIORAL ECOLOGIST. Teaching responsibilities will include teaching courses on animal behavior and human sexuality, and depending on training and experience other established undergraduate, including freshman, and graduate courses. Opportunities exist for new course development. Successful candidates will be broadly trained biologists with a Ph.D. Preference will be given to candidates who have demonstrated success in teaching and research involving undergraduates and the ability to teach a course that examines the ecological, evolutionary, and physiological basis of human sexuality. Submit materials to: **Dr. Michael P. Lombardo, Chair, Behavioral Ecologist Search Committee, Biology Department, Grand Valley State University, Allendale, MI 49401-9403, telephone: 616-331-2501, e-mail: lombardm@gvsu.edu, website: http://www.gvsu.edu/biology).**

Grand Valley State University is an Affirmative Action/ADA and Equal Opportunity Employer.

TWO ASSISTANT DIRECTORS
University of Notre Dame Environmental Research Center (UNDERC)

Two **ECOLOGISTS** with M.S. or Ph.D. degrees are sought to work with UNDERC Director **(Dr. Gary Belovsky)** in managing education, research, and workshop programs at either UNDERC-East or West. UNDERC-East is a 7,500 acre tract in the Upper Peninsula of Michigan and northern Wisconsin. UNDERC-West is a program in western Montana in partnership with the Confederated Salish and Kootenai Tribes.

These are 12-month Professional Specialist (M.S.) or nontenure-track faculty (Ph.D.) positions (three-year renewable contract) in the Department of Biological Sciences at the University of Notre Dame. Salary plus benefits are commensurate with education and experience. Presence at UNDERC-East (mid-May through September) or West (June through August) is required with the remainder of the year on campus. Housing at UNDERC-East and West is provided, as well as office and laboratory space at the UNDERC site and on campus.

Interested applicants should send curriculum vitae and a cover letter including description of research interests and teaching experience by October 15, 2006, to: **Dr. G. Belovsky, Department of Biological Sciences, P.O. Box 369, University of Notre Dame, Notre Dame, IN 46556-0369 (e-mail: belovsky.1@nd.edu).** Starting date for the position will be no later than April 1, 2007. *The University of Notre Dame is an Equal Opportunity/Affirmative Action Employer. Women and minorities are encouraged to apply.*

POSITIONS OPEN

ASSISTANT PROFESSOR, NEUROBIOLOGY
Department of Biological Studies
Bridgewater State College
Bridgewater, Massachusetts

Position: The position requires teaching introductory nonmajor biology courses and a seminar for the College's core curriculum, courses in neurobiology to augment the Department's offerings, and assisting in the development of an interdisciplinary program in neuroscience. Advising students and supervising original undergraduate research are also required.

Qualifications: The successful candidate must have an earned Ph.D. by June 2007, and excellent communication skills. Experience and/or strong interest in teaching biology to nonmajors, teaching courses in neurobiology, and supervising undergraduate research are required.

Preferred qualifications: Preference will be given to candidates with interests or experience in one or more of the following: bio-education outreach, teaching anatomy and physiology, postdoctoral experience, or grant experience.

Applicants should be strongly committed to excellence in teaching and advising, and to working in a multicultural environment that fosters diversity. They should also have an ability to use technology effectively in teaching and learning, the ability to work collaboratively, evidence of scholarly activity, and a commitment to public higher education.

To apply: Please visit our career site and apply online at **website: https://jobs.bridgew.edu.** Submit a letter of interest, curriculum vitae, a statement describing teaching and research interests, and names, addresses, and telephone numbers of three professional references for this position. Review of applications will continue until the position is filled. For more information about employment at Bridgewater State College, please visit our **website: http://www.bridgew.edu/HR/JobList/.**

Bridgewater State College is an Affirmative Action/Equal Opportunity Employer which actively seeks to increase the diversity of its workforce.

THE ENDOWMENT FOR SCHOLARS
in Biomedical Research at
The University of Texas Southwestern Medical Center

University of Texas (UT), Southwestern, is pleased to announce the continuation of the Endowed Program for Scholars in Biomedical Research. The Program, which is fully funded from private endowment, will provide $1,000,000 over four years to support the research activities of each new **ASSISTANT PROFESSOR** (tenure track) appointed to the Program; five will be appointed annually. Academic appointments and research space will be provided by individual medical school departments or research centers. Positions in both basic science and clinical departments are available. The goal of the program is to assure a successful beginning of the research careers of an ever-growing cadre of outstanding young investigators at UT Southwestern.

For detailed information about currently available faculty positions, please access our webpage **website: http://www8.utsouthwestern.edu/utsw/home/scholars/index.html.**

UT Southwestern is an Equal Opportunity Institution.

STAFF ASSOCIATE or SENIOR STAFF ASSOCIATE

Columbia University Department of Surgery is seeking a Staff Associate or Senior Staff Associate.

Requirements: Bachelor's degree in science discipline; at least four years of clinical experience in organ perfusion and preservation for transplantation; skills in sterile technique and gas sterilization procedures; skills in utilization of automated blood gas analyzers, including performance of and maintenance of quality control standards; working knowledge of Medtronic centrifugal cardiopulmonary bypass circuit.

Fax resumes to **Michele Silverthorne** at **fax: 212-305-6873.**

We take an Affirmative Action toward Equal Employment Opportunity.

POSITIONS OPEN

The University of Georgia

DIRECTOR
Georgia Sea Grant

The Georgia Sea Grant College Program seeks a new Director. Headquartered at the University of Georgia (UGA) in Athens, Georgia, Sea Grant (GSG) is part of the National Oceanic and Atmospheric Administration's (NOAA) National Sea Grant College Program, a network of 30 university-based programs in coastal and Great Lakes states. GSG has broad responsibilities in promoting marine research, education, and extension/advisory service throughout the State **(website: http://www.marsci.uga.edu/gaseagrant/).**

The Director oversees all Program activities, including research, outreach, education, program assessments, and fund raising. The successful candidate will have a Ph.D. or equivalent in a marine-related field; a relevant research focus; experience in administration and grant writing; broad knowledge of marine and aquatic science and resource issues at the national, regional, and state levels; strong communication skills and team-building skills; and the ability to work with diverse stakeholders. Prior experience with the Sea Grant College Program is desirable, but not required. The position will hold a tenure track/tenure appointment at the Associate/Full Professor level in the appropriate academic department.

Screening of applications will begin immediately and continue until a suitable candidate is selected. To insure full consideration, applications should be received by October 31, 2006. Applicants should send a cover letter, curriculum vitae, a summary of their professional interests, and contact information for three to five references to: **Dr. James T. Hollibaugh, Search Committee Chair, Department of Marine Sciences, Univeristy of Georgia, Athens, GA 30602-3636; e-mail: marsdir@uga.edu; telephone: 706-542-5868.** *UGA is an Affirmative Action/Equal Opportunity Institution.*

SYSTEMS BIOLOGIST
ASSISTANT PROFESSOR
Iowa State University

The Department of Genetics, Development, and Cell Biology **(website: http://www.gdcb.iastate.edu/)** invites applications for a tenure-track faculty position at the level of Assistant Professor. This position is part of a university-wide systems biology initiative and complements a dynamic interdepartmental group of biological and computational researchers. We seek highly qualified applicants from all backgrounds relevant to systems biology. Specific areas of interest include, but are not limited to: bioinformatics, computational biology, and modeling or system analysis of developmental, metabolic, or regulatory networks involving animals, plants, or microbes. The successful candidate will be expected to establish and maintain a vigorous, independent, extramurally funded research program, and to participate in undergraduate and graduate teaching. All applications must be submitted electronically at **website: http://www.iastatejobs.com** under vacancy identification 060856. To guarantee consideration the applications must be received by November 8, 2006.

The Department is sensitive to the needs of dual-career applicants and is committed to increasing diversity within the university community. Applications from women and members of under represented groups are strongly encouraged. Iowa State University is an Equal Opportunity/Affirmative Action Employer.

ASSISTANT PROFESSOR OF BIOLOGY, tenure track, Ph.D. in microbiology. Department of Biology, California State University, 9001 Stockdale Highway, Bakersfield, CA 93311-1099. See **website: http://www.csub.edu/Biology** for additional information. *California State University, Bakersfield is an Affirmative Action/Equal Opportunity Employer.*

POSITIONS OPEN

MICROBIOLOGIST

The Department of Biology at West Virginia University **(website: http://www.as.wvu.edu/biology/)** invites applications for a tenure-track position at the **ASSISTANT PROFESSOR** level in microbiology beginning August 2007. The successful candidate will develop an externally funded research program and demonstrate a commitment to excellence in both undergraduate and graduate education. Applicants must hold a Ph.D. or equivalent degree and have postdoctoral experience. Teaching assignment is expected to be primarily in upper division and graduate courses, including an upper level undergraduate course in microbial physiology. Significant contributions in teaching and research are required for retention and promotion.

We seek exceptional individuals in any field of microbiology, including those undertaking genomic, proteomic, or metabolomic research. The successful candidate should complement existing research programs in environmental biology, forensic biology, or model systems genetics. Additional interactions with members of the Davis College of Agriculture,Forestry and Consumer Sciences, the West Virginia University Health Sciences Center, and the National Institute for Occupational Safety and Health (NIOSH), are also available.

Qualified applicants should submit curriculum vitae, statements of research and teaching interests, representative publications, and three letters of recommendation to: **Microbiologist Search Committee Chair, Department of Biology, West Virginia University, P.O. Box 6057, Morgantown, WV 26506.** Review of applications will begin 15 November 2006.

For more information please contact us via **e-mail: jdwells@mail.wvu.edu,** or by **telephone: 304-293-5201, extension 31526.** *West Virginia University is an Equal Opportunity, Affirmative Action Employer, and does not discriminate on the basis of race, color, religion, sex, age, marital status, disability, veteran status, national origin, or sexual orientation.*

PHYSICAL OCEANOGRAPHER
Stanford University

The School of Earth Sciences seeks applications for a tenure-track faculty appointment at the **ASSISTANT or ASSOCIATE PROFESSOR** level in the area of physical oceanography. We are looking for a person with a commitment to excellence in both research and teaching. The successful applicant will be able to interface effectively with existing faculty members such as those currently using field, laboratory, and/or modeling-based approaches to study ocean ecology, biogeochemistry, climate variability, global change, coastal environmental engineering, and geomicrobiology. Excellent opportunities also exist for collaboration with the strong Marine Scientists within Stanford's Woods Institute and Hopkins Marine Station, and at nearby research institutes like the Carnegie Institution and the Monterey Bay Aquarium Research Institute.

Applications, including curriculum vitae, a statement outlining research and teaching experience and interests, and the names and addresses of three references should be sent in either electronic (PDF only) format or paper. Submit electronic applications to **e-mail: oceans-search@pangea.stanford.edu,** or if preferred, paper applications to:

Chair
Physical Oceanography Search Committee
School of Earth Sciences
Mitchell Building
Stanford, CA 94305-2215 U.S.A.

The deadline for applications is December 8, 2006. Additional information about School of Earth Sciences, Stanford University, can be found on its **website: http://pangea.stanford.edu/index.html.**

Stanford University has a strong institutional commitment to the principle of diversity. In that spirit, we particularly encourage applications from women, members of ethnic minorities, and individuals with disabilities.

International Institute of Molecular and Cell Biology in Warsaw **www.iimcb.gov.pl** the leading Polish research institute in the field of basic bio-medical sciences is seeking an

INDEPENDENT RESEARCHER

eligible to apply for prestigious **European Young Investigator Award** advertised by The Foundation for Polish Science at **http://www.fnp.eu/ang/programy/programy_euryi.html** and in *Science* of September 15, 2006. Interested scientists of any nationality, before submitting the application to the Foundation, in order to receive support of the International Institute of Molecular and Cell Biology as the host institution, should submit to IIMCB by November 15, 2006 at **SC@iimcb.gov.pl** full electronic application documentation requested by the Foundation.

The Institute offers to the award-winning candidate work in a scientifically stimulating environment of country-largest biomedical campus, furnished laboratory and office space in a modern building, possibility of selecting PhD students from leading Polish universities, access to the Institute's equipment, and fellowships for two PhD students for 5 years.

Extension of research activities of the group over the award period will be possible after positive evaluation according to IIMCB rules.

F A I R F I E L D U N I V E R S I T Y

Evolutionary Biologist

The Biology Department of the College of Arts and Sciences at Fairfield University announces a tenure-track assistant professor position in Evolutionary Biology with start date of September 1, 2007. We are seeking an Evolutionary Biologist using bioinformatics and computational biology to address questions of evolution in microbial, plant, or animal systems. Job requirements include: teaching undergraduates, maintaining an active research program that involves undergraduates, advising and mentoring students, and participating in departmental and university committees. Commitment to teaching excellence, responsiveness to student needs, and effective communication skills are expected.

Responsibilities will include participation in the teaching of the Biology department's introductory courses in general biology and genetics for Biology majors as well as an upper division course in the candidate's area of expertise. Candidates will also be expected to contribute to the University's science core curriculum. Applicants must possess a Ph.D. in Evolutionary Biology or a closely related discipline. Those with demonstrated excellence in undergraduate teaching, experience working with undergraduates in research, and post-doctoral research experience will be given special consideration. Applications from candidates with an appreciation of social and cultural diversity are encouraged. Salary and benefits at Fairfield University are highly competitive.

Qualified candidates should send a cover letter that addresses the above requirements. The application must include a curriculum vitae, graduate transcripts, a statement of teaching goals, a statement of research interests and goals (including the role of undergraduates and the potential for grant initiatives), selected reprints, and three letters of reference sent under separate cover. All application materials should be addressed to: **Dr. Glenn Sauer, Chair, Biology Department, Evolutionary Biologist Search, Fairfield University, Fairfield, CT 06824.** *Review of completed applications begins on October 15 and will continue until the position is filled.* Fairfield University is a comprehensive Jesuit university with an active and pluralistic faculty located in southern Connecticut, roughly 50 miles from New York City and minutes from New Haven CT.

Fairfield University is an Affirmative Action/Equal Opportunity Employer. Women, minorities, and persons with disabilities are strongly encouraged to apply.

Fairfield UNIVERSITY
Jesuit. Personal. Powerful.

Visit our website at www.fairfield.edu

The Ottawa Institute of Systems Biology
Tenure-track positions in System Biology
Ottawa Institute of Systems Biology, Faculty of Medicine
University of Ottawa

The Ottawa Institute of Systems Biology (**www.oisb.ca**) is seeking candidates for junior and senior tenure-track faculty positions. Our purpose is to advance systems biology methodologies for studying cellular functions and diseases, with a particular interest in cancer and neurobiology. Candidates with research interests in the application of high-throughput technologies such as genomics, proteomics, glycomics, lipidomics, RNAomics, and chemical biology are encouraged to apply. We are also seeking candidates interested in bridging structural biology (x-ray, NMR, and in-silico) with systems biology, in bioinformatics, and in computational biology. All candidates will be considered for the Canada Research Chair program. The University of Ottawa provides a vibrant bilingual research environment in the Capital of Canada. Qualified applicants are invited to submit a letter of application, curriculum vitae, a statement of research interests, and arrange to have three letters of recommendation sent to **Dr. Daniel Figeys** at **sysbio@uottawa.ca**.

All qualified candidates are encouraged to apply; however, Canadian citizens and permanent residents will be given priority. Equity is a University of Ottawa policy; women, aboriginal people, members of visible minorities and persons with disabilities are especially welcome to apply. Evaluation of applications will continue until available positions are filled. We thank all applicants who apply. Only those selected for an interview will be contacted.

F A I R F I E L D U N I V E R S I T Y

Developmental Geneticist

The Department of Biology at Fairfield University announces a tenure-track position at the Assistant Professor level in the area of Developmental Genetics, to begin fall 2007. Candidates should have expertise in genetics and will be expected to develop a research program in developmental biology and/or genetics in animals. Job requirements include teaching undergraduates, maintaining an active research program involving undergraduates, advising and mentoring students, and participating in departmental and university committees. Commitment to teaching excellence, responsiveness to student needs, and effective communication skills are expected.

Responsibilities will include participation in the teaching of the Biology department's introductory courses in general biology and genetics for Biology majors, as well as an upper division course in developmental biology. Candidates will also be expected to contribute to the University's science core curriculum. Candidates must possess a Ph.D. in genetics or a closely related discipline. Those with demonstrated excellence in undergraduate teaching, experience working with undergraduates in research, and post-doctoral research experience will be given special consideration. Applications from candidates with an appreciation of social and cultural diversity are encouraged. Salary and benefits at Fairfield University are highly competitive.

Qualified candidates should send a cover letter that addresses the above requirements. The application must include a curriculum vitae, graduate transcripts, a statement of teaching goals, a statement of research interests and goals (including the role of undergraduates and the potential for grant initiatives), selected reprints, and three letters of reference sent under separate cover. All application materials should be addressed to: **Dr. Glenn Sauer, Chair, Biology Department, Developmental Genetics Search, Fairfield University, Fairfield, CT 06824.** *Review of completed applications will begin on November 1st, and will continue until the position is filled.* Fairfield University is a comprehensive Jesuit university with an active and pluralistic faculty located in southern Connecticut, roughly 50 miles from New York City and minutes from New Haven CT.

Fairfield University is an Affirmative Action/Equal Opportunity Employer. Women, minorities, and persons with disabilities are strongly encouraged to apply.

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POSITIONS OPEN

MICROBIAL ECOLOGIST TENURE-TRACK FACULTY POSITION
University of Toronto at Mississauga

The University of Toronto at Mississauga (UTM), Department of Biology, invites applications for a tenure-track faculty position in microbial ecology at the level of **ASSISTANT PROFESSOR,** effective July 1, 2007.

The area of specialization is open with preference to studies of biodiversity, community dynamics, and biogeochemical activities.

Opportunities exist for collaboration with Cell and Molecular Biologists with those interested in the evolution and ecology of both terrestrial and aquatic systems. Depending on the area of speciality, a Microbial Ecologist may interact with the biologically oriented Chemists and Wetland Geographers interested in toxicity.

The successful applicant will have a Ph.D. and preferably postdoctoral experience, an outstanding academic record, and demonstrated excellence in research and teaching. The appointee will be expected to build an active, externally funded and internationally recognized research program and to contribute to the education and training of undergraduate and graduate students. Salary will be commensurate with qualifications and experience.

Applicants should provide curriculum vitae, a statement of teaching philosophy and interests, an outline of their proposed research, and should arrange to have three confidential letters of recommendation sent on their behalf to: **Professor Robert Reisz, Chair, Department of Biology, University of Toronto at Mississauga, Mississauga, Ontario, Canada L5L 1C6. E-mail: rreisz@utm.utoronto.ca.** Closing date for submissions is December 1, 2006. For more information on the Department go to **website: http://www.utm.utoronto.ca/~w3bio/homepage/.**

The University of Toronto is strongly committed to diversity within its community and especially welcomes applications from visible minority group members, women, Aboriginal persons, persons with disabilities, members of sexual minority groups, and others who may contribute to the further diversification of ideas.

All qualified candidates are encouraged to apply; however, Canadians and permanent residents will be given priority.

The Department of Psychology at the University of Wisconsin, Madison, seeks to appoint an outstanding **SCHOLAR**, rank open, engaged in research in experimental psychopathology/clinical psychology. We seek candidates with high-impact, novel research programs. Our emphasis is on scholarly distinction without regard to specific research focus. Applications should be received by November 1, 2006, for full consideration, but applications will be accepted until the position is filled. See our **website:http://psych.wisc.edu** to learn about the Department and other outstanding research units. Ph.D. required prior to start of appointment. Applicants should send curriculum vitae, reprints or preprints, and statement of research and teaching interests to: **New Personnel Committee, Clinical Position, Department of Psychology, University of Wisconsin-Madison, 1202 West Johnson Street, Madison, WI 53706-1611.** Three letters of reference should be sent for junior candidates. Tenured applicants should submit a list of three references whom we may contact. Unless confidentiality is requested in writing, information regarding applicants must be released upon request. Finalists cannot be guaranteed confidentiality. *The University of Wisconsin, Madison, is an Affirmative Action/Equal Opportunity Employer. Women and minorities are encouraged to apply.*

LANDSCAPE ECOLOGIST. Department of Wildlife Ecology and Conservation **(website: http://www.wec.ufl.edu),** University of Florida, U.S.A. We are hiring an **ASSISTANT PROFESSOR,** with a 60 percent research and 40 percent teaching split in a 12-month tenure-track position. Review of applications begins 14 November 2006. See **website: http://www.wec.ufl.edu/lej.pdf** for application procedures and full position description.

POSITIONS OPEN

ASSISTANT/ASSOCIATE PROFESSOR
Plant Evolutionary Genetics

The School of Biological Sciences at Washington State University in Pullman, Washington, invites applications for a full-time tenure-track position in plant evolutionary genetics to begin August 2007, at the **ASSISTANT or ASSOCIATE PROFESSOR** level. Applicants should have ability and potential for outstanding teaching and for maintaining a strong empirical research program in plant evolutionary genetics, focusing on questions that complement our faculty's strengths in population and ecological genetics, evolutionary ecology, molecular evolution, systematics, ecology, and physiology. Candidates pursuing rigorous, theory-driven empirical research on plant evolutionary genetics using sophisticated quantitative skills are particularly encouraged to apply, as are individuals who are effective communicators with broad knowledge of plant biology and interests in collaborative research and training. Required qualifications include an earned doctorate at time of application, a record of research accomplishment commensurate with rank in plant evolutionary genetics, and a commitment to teaching excellence in undergraduate and graduate courses. Successful candidates will be expected to develop and maintain a vigorous, independent research program supported by extramural funding, train graduate and undergraduate students, participate in graduate and undergraduate teaching including a graduate course in population genetics, and shared responsibilities for undergraduate courses in general genetics or evolution, and advance the College's commitment to diversity and multiculturalism.

To apply, send a letter of application addressing qualifications, curriculum vitae, statements of research and teaching interests, and a list of names, addresses, and telephone numbers of at least three references. Arrange for at least three letters of reference to be sent directly to the **Search Committee.** These letters of reference should clearly address your research potential, teaching and communication skills. Send all materials by November 13, 2006, to: **Plant Evolutionary Genetics Search Committee, c/o Linda Larrabee, School of Biological Sciences, P.O. Box 644236, Pullman, WA 99164-4236, e-mail: larrabee@wsu.edu, telephone: 509-335-5768, fax: 509-335- 3184.** Full notice of vacancy can be viewed at **website: http://www.sci.wsu.edu/sbs/index.php3.** *Equal Employment Opportunity/Affirmative Action/ADA.*

NATIONAL UNIVERSITY OF SINGAPORE
Department of Chemical and Biomolecular Engineering

The Department of Chemical and Biomolecular Engineering at National University of Singapore invites applications for **TENURE-TRACK FACULTY** positions at all levels. The Department is one of the largest internationally with excellent in-house infrastructure for experimental and computational research. A Ph.D. in chemical engineering or related areas and a strong research record with excellent publications are required. Please refer to **website: http://www.chbe.nus.edu.sg/** for more information on the areas of interest and for application details. Applicants should send full curriculum vitae (including key publications), a detailed research plan, a statement of teaching interest, and a list of names of at least three references to: **Professor Raj Rajagopalan, Head of Department (Attention: Ms. Nancy Chia, e-mail: nancychia@nus.edu.sg).**

ASSISTANT/ASSOCIATE PROFESSOR, PHARMACEUTICAL SCIENCES, LOMA LINDA UNIVERSITY SCHOOL OF PHARMACY. Ph.D. in pharmacology, medicinal chemistry, or related biomedical sciences for teaching physiology, biochemistry, molecular biology, or pharmacology. Record of excellence in teaching, service, and research. Forward curriculum vitae, letter of interest, and reference information to: **John Krstenansky, 11262 Campus Street, Loma Linda University, Loma Linda, CA 92350 (e-mail:jkrstenansky@llu.edu).**

POSITIONS OPEN

ASSISTANT PROFESSOR OF GEOCHEMISTRY
Washington University in St. Louis

Washington University in St. Louis invites applications for a tenure-track Assistant Professor position in low-temperature geochemistry to begin in fall 2007. We seek candidates who apply modern, quantitative techniques and demonstrate or show promise of excellence in both teaching and research. Of particular interest are candidates with expertise in organic, aqueous, or biogeochemistry. Research areas may include, but are not limited to, the degradation and preservation of carbonaceous materials in soils and sediments, isotope fractionations, and variations through geologic time, remnant complex carbon molecules in the rock record as markers of past biological processes, and the role of past or present microorganisms in the carbon or other elemental cycles. Candidates must have been awarded the Ph.D. at time of appointment and should send curriculum vitae, statement of future research interests, and names and contact information for at least four references to: **Jan P. Amend, Search Committee Chair, Department of Earth and Planetary Sciences, Washington University, C/B 1169, One Brookings Drive, St. Louis, MO 63130,** or via **e-mail: gcfacsearch @levee.wustl.edu.** Applications will be considered until the position is filled, but priority will be given to those received by November 30, 2006. *Women and minorities are encouraged to apply. Washington University is an Equal Opportunity/Affirmative Action Employer. Employment eligibility verification required upon employment.*

BIOCHEMISTRY. Baylor University announces a faculty position, **ASSISTANT PROFESSOR** of biochemistry, beginning fall 2007. The Department is housed in a new $103 million science facility, part of an exciting plan of growth at the University. Any area of biochemistry will be considered. The successful candidate will enjoy many opportunities for teaching and research in a dynamic environment that fosters interdisciplinary collaboration. Requirements: Ph.D. in biochemistry, chemistry, or closely related field; commitment to exemplary teaching at both the undergraduate and graduate level; and potential for a vigorous, independent, externally funded research program. Postdoctoral experience is desirable. Send letter of application, full curriculum vitae, summary of future research plans, estimate of startup costs, undergraduate and graduate transcripts, and three letters of reference to: **Chair, Search Committee, Department of Chemistry and Biochemistry, One Bear Place #97348, Baylor University, Waco, TX 76798-7348. E-mail: mary_lynn_trawick@baylor.edu.** Applications will be reviewed beginning September 4, 2006, and will be accepted until the position is filled. To ensure full consideration, your application must be completed by October 25, 2006. Baylor is a Baptist university affiliated with the Baptist General Convention of Texas. *As an Affirmative Action/Equal Employment Opportunity Employer, Baylor encourages minorities, women, veterans, and persons with disabilities to apply.*

BIOLOGY POSITION

The Department of Integrative Biology at Brigham Young University (BYU) has a permanent **FACULTY** position available starting as early as January 2007. Responsibilities include teaching large sections of a general education biology course and developing undergraduate and graduate courses in area of expertise. Ph.D. in a biological discipline or biological science education, postdoctoral work, and teaching experience required. Development of an externally funded research program expected. Send one-page letter of interest and two-page curriculum vitae to: **Dr. Rex G. Cates, 425 WIDB, Integrative Biology, Brigham Young University, Provo, UT 84602.** Closing date is 15 October 2006.

BYU, an Equal Opportunity Employer, is sponsored by the Church of Jesus Christ of Latter-day Saints and requires observance of Church standards. Preference is given to members in good standing of the sponsoring Church.

BME Faculty Position Available

Tufts University, Department of Biomedical Engineering – is seeking candidates for a faculty appointment at the **Assistant, Associate**, or **Full Professor** level. Rank will be determined by experience and accomplishments. Preference will be given to candidates whose research interests involve regenerative medicine. The successful candidate will join an active Ph.D.-granting Department and must demonstrate the potential to develop an outstanding, internationally recognized research program, excel in teaching, and develop strong interschool collaborations. Teaching responsibilities include graduate and undergraduate courses. A Doctorate is required and postdoctoral experience is desirable. Additional information about the department can be found at **ase.tufts.edu/biomedical/**.

Interested applicants should send their Curriculum Vitae, cover letter, research plan, and names of three references to: **Faculty Search Committee, Department of Biomedical Engineering, Tufts University, 4 Colby Street, Medford, MA 02155**. Evaluation of candidates will begin at the end of October 2006 and continue until the position is filled.

Tufts University is an Affirmative Action/ Equal Opportunity Employer. We are committed to increasing the diversity of our faculty. Applications from women and members of underrepresented groups are strongly encouraged.

Chair of Department of Biomedical Engineering
The University of Texas

The University of Texas seeks an experienced and imaginative academic leader with a distinguished record of accomplishments in research to fill the position of Chair of the Department of Biomedical Engineering. The Department has recently been formed by joining the existing BME Department at the University of Texas at Austin (UTA) with the University of Texas M.D. Anderson Cancer Center (UTMDACC) and the University of Texas Health Science Center at Houston (UTHSC-H). This new department combines the strengths of premier engineering and medical institutions to create an extraordinary center of excellence in biomedical engineering with exceptional resources. Innumerable opportunities are available to lead and develop new research and educational programs in biomedical engineering among these institutions.

The BME Chair has responsibility for leading the Department across all three institutions with the assistance of a deputy chair at each institution and a senior level interinstitutional administrator. The Department consists of more than 30 primary faculty, a majority of whom are at the junior level, and more than 50 affiliated faculty. The Department has a very strong program of externally funded research. Excellence in education is a high priority of the Department, and many faculty have won teaching awards. The undergraduate student body numbers approximately 450, and the graduate students are about 100. A primary responsibility of the Chair is to oversee the undergraduate program which is administrated fully in Austin. An accreditation review is scheduled for September 2006. A new BME building of 141,000 ft^2 is under construction with a completion anticipated in mid-2008. Dedicated facilities for the Department exist in Houston, and space in a new building has been approved. The Chairman reports to an Inter-institutional Administrative Committee consisting of the Dean of Engineering at UT Austin, the Chief Academic Officer at UTMDACC, and the Executive Vice President of Academic Affairs at UTHSC-H.

The successful candidate will have a Ph.D., M.D. or both, with academic achievements commensurate with appointment as a full professor with tenure, a highly respected record of research in biomedical engineering, and significant administrative experience. The candidate must be a strong leader, a capable motivator, and be able to develop the talents of others in support of the Department's mission. It is expected that the Chair will be an active participant in research and teaching activities.

A curriculum vitae, names of 3 to 5 references (with titles, affiliations and complete contact information), and a brief synopsis of experience and career goals should be e-mailed to **burks@mail.utexas.edu**.

Women and minority candidates are encouraged to apply. The University of Texas is an EO/AA employer. M/F/D/V. This is a security sensitive position and thereby subject to Texas Education Code §51.215. A background check will be required for the final candidate.

LOYOLA MARYMOUNT UNIVERSITY
Chair for the Department of Biology

The Department of Biology welcomes applications for the position of Department Chair and Professor, with duties to begin in Summer 2007. This vibrant department offers Bachelor of Science and Bachelor of Arts degrees in Biology, and consists of four professors, four associate professors, four assistant professors, and three staff members. The successful candidate will be chosen from associate or full professor applicants. Candidates should possess demonstrated excellence in teaching and research; prior administrative experience is desirable. The area of expertise ideally will supplement existing departmental strengths. The chair is expected to teach in lower and upper division courses and to maintain an active research program involving undergraduates. In addition, the Chair will be involved in developing select interdisciplinary graduate programs at the master's level consistent with the College of Science and Engineering's Strategic Plan.

Loyola Marymount University (LMU), founded in 1911, is a comprehensive university in the mainstream of American Catholic higher education. Located on the west side of Los Angeles overlooking the Pacific, LMU is one of the nation's 28 Jesuit colleges and universities and five Marymount institutions. It serves 5,400 undergraduates and over 2,500 graduate students in the Colleges/ Schools of Science and Engineering, Liberal Arts, Business Administration, Communication and Fine Arts, Film and Television, Education, and Law.

Loyola Marymount University seeks professionally outstanding applicants who value its mission and share its commitment to academic excellence, the education of the whole person, and the building of a just society. (Visit **www.lmu.edu** for more information.)

Please submit a letter of application, a current vitae, statements of teaching and research philosophies, three letters of reference and a statement of vision for a biology department dedicated to the education of undergraduates in a college climate fostering interdisciplinary programs. Review of applications will begin on **October 20, 2006**. Materials should be sent to: **Prof. James Landry, Chair of Search Committee, Department of Natural Science, Loyola Marymount University, 1 LMU Drive, MS 8160, Los Angeles, CA 90045-2659.**

LMU is an Equal Opportunity Institution actively working to promote an intercultural learning community. Therefore, we encourage applications from underrepresented groups.

HUMAN MICROBIAL PATHOGENESIS
FACULTY POSITIONS – OPEN RANK
CENTER FOR MOLECULAR AND TRANSLATIONAL HUMAN INFECTIOUS DISEASE RESEARCH

THE METHODIST HOSPITAL RESEARCH INSTITUTE

The Methodist Hospital Research Institute (TMHRI) at The Methodist Hospital (TMH) in Houston, Texas, seeks several exceptional scientists studying the molecular basis of human microbial pathogenesis. Individuals who currently lead multiple-PI teams, and collaborating PIs who desire to co-locate also will be considered.

TMHRI has academic partnerships with the University of Houston and Weill Medical College of Cornell University in New York City. TMH has entered an unprecedented expansion phase that includes building a 340,000 SF state-of-the-art research building with bio-containment, non-human primate, and molecular imaging facilities, and a 600,000 SF ambulatory care building, both designed to foster interdisciplinary collaborative research.

We are interested in candidates using new technologies to study molecular events occurring at the host-pathogen interface.

Successful applicants will be responsible for establishing or expanding nationally recognized, externally funded research programs.

Applicants must have an advanced degree (PhD, DVM, MD, or MD/PhD). Successful applicants will receive an outstanding recruitment package. Interested individuals should send via e-mail a curriculum vitae; description of research interests, future directions, and grant funding information; and the names of at least three references to:

James M. Musser, M.D., Ph.D.
c/o Ms. Irene Harrison, E-mail: iaharrison@tmh.tmc.edu
Co-Director and Executive Vice President
The Methodist Hospital Research Institute
6565 Fannin St., Mail Stop B490, Houston, TX 77030
EOE

Methodist The Methodist Hospital Research Institute

CORNELL UNIVERSITY Joan and Sanford I. Weill Medical College

POSITIONS OPEN

ANALYTICAL CHEMISTRY or MATERIALS CHEMISTRY

The Department of Chemistry at the University of Michigan invites applications for an anticipated position in the area of analytical chemistry or materials chemistry at the rank of **ASSISTANT or ASSOCIATE PROFESSOR** with a proposed start date of September 1, 2007. This would be a University-year appointment (nine months academic salary with three months research supported salary.) Applications in all areas of analytical chemistry are encouraged, including but not limited to, bioanalytical, mass spectrometry, sensors, separations, and spectroscopy. Specific areas in materials chemistry may include but will not be limited to polymer synthesis and applications, supramolecular chemistry, inorganic and organic biomaterials, sensors, and optical or electronic materials. Interdisciplinary graduate programs at Michigan available for research collaborations include applied physics, biophysics, and macromolecular science and engineering. Detailed information regarding the electronic application process and required materials is available online at **website: http://www.chem.lsa.umich.edu/chem/facultyrecruit/.** The position will remain open until filled but preference will be given to applicants who have submitted all requested materials prior to October 15, 2006. Information about the Chemistry Department is available on the **website: http://www.umich.edu/~michchem.** Questions about the applications process should be sent to **e-mail: chemfac05@umich.edu.** *Women and minorities are encouraged to apply. The University of Michigan is supportive of the needs of dual-career couples and is a nondiscriminatory, Affirmative Action Employer.*

FACULTY POSITIONS (TWO) IN BIOLOGY

York College of the City University of New York invites applications for two tenure-track positions at the **ASSISTANT PROFESSOR** level in genetics/bioinformatics and in plant physiology/evolution to begin September 1, 2007. Qualifications include a Ph.D. with postdoctoral experience and evidence of excellence in teaching. Instructional responsibilities include lecture and laboratory courses in area of expertise as well as other major or nonmajor courses as needed. Candidates must demonstrate a strong interest and commitment to undergraduate teaching and the capability of developing and maintaining an active research program supported by external funding. The academic program and instructional and research equipment available at York College can be found at **website: http://natsci.york.cuny.edu.** Applicants should submit a cover letter, curriculum vitae, statements of research and teaching experience, and the names and contact information of three professional references to: **Dr. Margaret MacNeil, Biology, York College/CUNY, 94-20 Guy R. Brewer Boulevard, Jamaica, NY 11451.** The application deadline is October 23, 2006. *Equal Employment Opportunity/Affirmative Action/ADA/IRCA.*

DIVISION DIRECTOR
Illinois Natural History Survey (INHS)

Director of the Division for Ecology and Conservation Sciences is a Senior Administrator at the level of Professional or Senior Professional Scientist (full-time, state funded). The Director provides leadership, vision, and direction on significant initiatives, research projects, and outreach programs within the Division and manages staff and budgets. Applicants must have a broad scientific background in natural resources, conservation science, or basic or applied ecology; demonstrated ability to effectively lead and manage staff; and a strong interest in public/government service. A Ph.D. in biological sciences with minimum of eight years of relevant experience is required. Located on the University of Illinois Urbana/Champaign campus. For a complete position description and application requirements visit our **website: http://www.inhs.uiuc.edu/opportunities.**

POSITIONS OPEN

TENURE-TRACK FACULTY POSITION
Oral Biology Department
Indiana University School of Dentistry

A full-time, tenure-track/tenured position is available now at the **ASSISTANT/ASSOCIATE PROFESSOR** level in the areas of biochemistry, cell biology, molecular biology, or biofilms in the Department of Oral Biology at Indiana University (IU) School of Dentistry. Other areas of specialization may be considered. For information on the Department please visit the IU School of Dentistry, Department of Oral Biology Home Page **website: http://www.iusd.iupui.edu/Depts/OB/.**

The successful candidate must have a Ph.D. or Ph.D./D.D.S. and demonstrated expertise in the candidate's area of research. Postdoctoral experience is preferred. Applicants at the level of Associate Professor are expected to have a record of extramural funding and peer–reviewed publications.

The School of Dentistry provides a modified problem-based learning (PBL) dental curriculum. Candidates will be expected to participate in PBL tutorial groups and courses in molecular cell biology, systems approach to biomedical sciences, and appropriate graduate level courses.

A letter of application, a statement of present and future research interests, curriculum vitae, and the names of three references should be sent electronically to: **Dr. Chris H. Miller, Office of Academic Affairs,** at **e-mail: chmille@iupui.edu.** Review of applications will begin on October 1, 2006, and continue until an acceptable candidate is identified. *Indiana University is an Equal Employment Opportunity / Affirmative Action Employer.*

CHEMISTRY

The Department of Chemistry and the Life Sciences Institute (LSI) at the University of Michigan invite applications for an anticipated position at the rank of **ASSISTANT PROFESSOR or ASSOCIATE PROFESSOR** in the field of chemistry with a proposed start date of September 1, 2007. Any area of chemistry that overlaps with life sciences is of interest including, but not limited to, bio-analytical, biomaterials, and bio-organic. The successful candidate's laboratory will be located in the LSI, a scientific enterprise at the University of Michigan dedicated to opening new scientific paths by blending diverse research talents in a state-of-the-art collaborative facility. Candidates are expected to develop an internationally recognized program of scholarly research and to excel in teaching at undergraduate and graduate levels. Detailed information regarding the electronic application process and required material is available online at **website: http://www.chem.lsa.umich.edu/chem/facultyrecruit/.** The position will remain open until filled but preference will be given to applicants who have submitted all requested materials prior to October 15, 2006. Information about the Chemistry Department and LSI is available online **(websites:http://www.umich.edu/~michchem; www.lifesciences.umich.edu/institute/).** Questions about the application process should be sent to **e-mail: chemfac06@umich.edu.** *Women and minorities are encouraged to apply. The University of Michigan is supportive of the needs of dual-career couples and is a nondiscriminatory, Affirmative Action Employer.*

RESEARCH ASSOCIATE sought by the Alfred I. duPont Hospital for Children/Nemours Foundation for the Molecular Genetics, Cellular and Tissue Transplantation Laboratory in Wilmington, Delaware. Must have Ph.D. in biophysics or biochemistry with minimum of two years of postdoctoral experience. Requirements include the ability to perform yeast 2 hybrid screening, protein phosphatase-1 activity and kinetic assays, as well as knowledge of targeting of phosphorylated proteins inhibiting the HIF complex in the oxygen-sensing pathway in human cells, gained through prior experience or thesis research. Send resume to **A.M. Riddle, M.B.A., Certified Healthcare Executive, Administrator for Nemours Biomedical Research, A.I. duPont Hospital for Children, P.O. Box 269, Wilmington, DE 19899.**

POSITIONS OPEN

IMMUNOLOGY FACULTY POSITION

The Department of Microbial and Molecular Pathogenesis is recruiting a tenure-track **IMMUNOLOGIST** at the **ASSISTANT PROFESSOR** level. Investigators whose research complements existing strengths in host-pathogen interactions and vaccinology are encouraged to apply. State-of-the-art animal care, biocontainment (BSL-3), and other core facilities are available within the Department and elsewhere on campus. Candidates are expected to establish an independent, extramurally funded research program, to teach immunology to professional and graduate students, and to participate in an NIH predoctoral training grant in host-pathogen interactions. Numerous opportunities exist for collaborations in basic sciences as well as interdisciplinary programs with the College of Veterinary Medicine and other components of Texas A&M University and the Health Science Center, including the Texas Institute for Genomic Medicine. Appointments are for 12 months with a competitive startup package and laboratory space. Applicants should provide curriculum vitae, a statement of research interests, and have letters submitted by three referees. These materials should be sent online **(e-mail: dmcmurray@tamu.edu)** and by post to: **Professor David N. McMurray, Search Committee Chair, Department of Microbial and Molecular Pathogenesis, College of Medicine, Texas A&M University System Health Science Center, College Station, TX 77843-1114.** For more information, visit our **website: http://medicine.tamhsc.edu/basic_sciences/mmp/.**

The Texas A&M University System Health Science Center is an Affirmative Action/Equal Opportunity Employer and encourages applications from women and minorities.

COMPUTATIONAL BIOCHEMISTRY FACULTY POSITION
Kansas State University

The Department of Biochemistry at Kansas State University **(website: http://www.ksu.edu/bchem)** invites applications for a tenure-track position at the **ASSISTANT PROFESSOR** level, to begin in August 2007. Applicants should have a Ph.D. or equivalent degree and postdoctoral experience in an area of computational biochemistry. Preference will be given to applicants with research interest and experience in areas that complement and enhance the existing programs in the Department. The Department seeks individuals who will sustain a strong, extramurally funded research program and excel in teaching a diverse population in the undergraduate and graduate programs in biochemistry. The position will include a competitive salary and startup package. For additional information about the Department and the position, see **website: http://www.ksu.edu/bchem.** Applicants should submit curriculum vitae, statement of research and teaching interests, reprints, and three letters of reference to: **Michael R. Kanost, Head, Department of Biochemistry, 141 Chalmers Hall, Kansas State University, Manhattan, KS, 66506-3902.** Review of applications will begin on October 20, 2006, and continue until the position is filled. *Kansas State University is an Equal Opportunity Employer and actively seeks diversity among its employees.*

FACULTY POSITION IN CELL BIOLOGY

The Biology Department of Indiana University Southeast (IUS) invites applications from qualified Ph.D.s for a tenure-track **ASSISTANT PROFESSOR** position in cell biology to begin August 2007. Postdoctoral experience is preferred. Successful candidate will teach cell biology, introductory biology, and other advanced courses, and is expected to pursue a research program. Research space, start-up funds, release time for research, and competitive salary and benefits will be provided. For detailed information and directions to apply visit **website: http://www.ius.edu/HR/.** Postmarked deadline: November 15, 2006. *IUS is an Affirmative Action/Equal Opportunity Employer.*

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Advertising Supplement

Read this special ad supplement devoted to stem cell research career opportunities in the **6 October issue of *Science*.**

Find stem cell research jobs and other career resources online at **www.sciencecareers.org.**

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e-mail: danderso@aaas.org

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phone: +81 (0) 52 789-1860
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PENNSTATE
University Park

Head, Department of Biochemistry and Molecular Biology

Penn State is seeking an individual with an outstanding research record and excellent interpersonal skills who can provide energetic and creative leadership for its Department of Biochemistry and Molecular Biology. The Department has 38 faculty members with interests that include gene regulation, genomics and bioinformatics, biophysics and structural biochemistry, microbiology, virology, cell biology and developmental biology. A number of additional faculty appointments are anticipated over the next few years. The research programs in the Department are currently supported by annual grant awards exceeding $12 million. The Department has vigorous undergraduate and graduate educational programs and participates in a number of interdisciplinary graduate programs. The Department also plays a major role in the broader life sciences community in the Eberly College of Science and across Penn State. Further detailed information may be found at the Department's Web site at http://www.bmb.psu.edu. The position is available for Fall 2007. Credentials appropriate to the rank of tenured full professor are required. Review of applications and nominations will begin October 1, 2006 and will continue until the position is filled. Applications, including curriculum vitae and the names of three references, and nominations may be submitted via email to mlbl@psu.edu or mailed to:

BMB Head Search Committee
Eberly College of Science, 517 Thomas Building
The Pennsylvania State University
University Park, PA 16802

Penn State is committed to affirmative action, equal opportunity and the diversity of its workforce.

PENN STATE ***Making Life Better***

AUBURN UNIVERSITY
COLLEGE OF SCIENCES AND MATHEMATICS

Chair, Department of Biological Sciences

The Department of Biological Sciences at Auburn University invites applications and nominations for the position of Chair. Biological Sciences is an integrative department within the College of Sciences and Mathematics with expertise in a diverse array of biological disciplines offering M.S. and Ph.D. degrees to ~100 graduate students and B.S. degrees to ~550 undergraduate majors. More information about the department is available at **http://www.auburn.edu/biology**. A nationally recognized program of excellence in biological sciences has been identified as a University priority.

Applicants must have a Ph.D. in Biological Sciences or closely related life sciences discipline, as well as a record of academic excellence consistent with appointment as Full Professor. Dynamic leadership and strong interpersonal skills are also required. The Chair functions as an administrative scholar, providing visionary leadership to carry the Department of Biological Sciences forward in the areas of research, teaching, and outreach. The selected candidate must be eligible for employment in the U.S. at the date of appointment and be able to communicate effectively in English.

Applicants should submit a detailed curriculum vitae, transcripts, representative reprints, a statement of administrative philosophy, personal teaching and research goals, and the names and contact information of at least three references. Applications should be sent to: **Dr. Charles Savrda, Biological Sciences Search Committee Chair, Dept. of Biological Sciences, 101 Life Sciences Building, Auburn University, AL 36849-5407**, or electronically as PDFs to **bioscichairsearch@auburn.edu**. Review of applications will begin **November 1, 2006**.

AUBURN UNIVERSITY IS AN AFFIRMATIVE ACTION/EQUAL OPPORTUNITY EMPLOYER.
Women and minorities are encouraged to apply.

Mathematical Biology Faculty
LOYOLA MARYMOUNT UNIVERSITY

The College of Science and Engineering seeks candidates for a Presidential Professorship in Mathematical Biology. Candidates must have a distinguished record in teaching and research and a clear vision for providing leadership in interdisciplinary educational and research programs in Mathematical Biology. The ideal candidate will receive a joint appointment in Biology and Mathematics at the rank of Professor. Our College's faculty have a variety of current and emerging research interests, including bioinformatics, coding theory, dynamics, ecology and evolution, epidemiology, genomics, knot theory, modeling, proteomics, and probability and statistics. The individual we are seeking will broaden and complement our current interests and expertise. LMU currently maintains an individualized studies undergraduate degree in Biomathematics, and we are actively developing a formal major. Our College currently participates in two REU programs, with more under development. The successful candidate will provide leadership not only in the undergraduate degree programs and any future initiatives but also the recruitment of additional faculty to strengthen interdisciplinary interactions among departments in the College.

Requirements for the position include a Ph.D. in Biology, Mathematics, or a relevant, related discipline. Applicants are requested to send a letter of application, curriculum vitae, vision statement for the position, and three letters of reference. Review of applicants is ongoing and will continue until the position is filled. Materials should be sent to: **Biomathematics Search Committee, Department of Mathematics, UH 2700, Loyola Marymount University, 1 LMU Drive, Los Angeles, CA 90045-2659**. For additional information, contact **Dr. Ben Fitzpatrick, (310) 338-7892**. To learn more about LMU and the College, visit **www.lmu.edu** and **cse.lmu.edu**.

Loyola Marymount, a comprehensive university in the mainstream of American Catholic higher education, seeks professionally outstanding applicants who value its mission and share its commitment to academic excellence, the education of the whole person, and the building of a just society. LMU is an Equal Opportunity Institution actively working to promote an intercultural learning community. Women and minorities are encouraged to apply.

POSITIONS OPEN

FACULTY POSITION IN MICROBIOLOGY
Molecular Genetics, Biochemistry, and Microbiology
University of Cincinnati

The Department of Molecular Genetics, Biochemistry and Microbiology seeks to fill a tenure-track faculty position at any level **(ASSISTANT/ASSOCIATE/FULL PROFESSOR).** Successful candidates will have already established a highly competitive independent research program or be able to develop such a program.

We are particularly interested in microbial pathogenesis including emerging infectious diseases, mechanisms of disease resistance, and analysis of microbial genomes, signal transduction in viral/host interactions.

The Department currently has 24 full-time faculty and active graduate and postdoctoral programs. The College has a state of the art BL3 containment facility, and core facilities support advanced microscopy (electron and confocal) and imaging, gene microarray analysis, proteomics, informatics, and production of transgenic and knockout mice. Our Departmental structural biology program includes both nuclear magnetic resonance and X-ray crystallography.

For further information, please see **website: http://www.molgen.uc.edu/logic/.** Applicants should submit curriculum vitae, a brief description of research, and the names of three qualified references to: **Jerry B. Lingrel, Ph.D., Professor and Chair, Department of Molecular Genetics, Biochemistry and Microbiology, P.O. Box 670524, Cincinnati, OH 45267-0524.**

FACULTY POSITION IN CARDIOVASCULAR BIOLOGY

The Department of Molecular Genetics, Biochemistry and Microbiology is recruiting for a tenure-track faculty position at the **ASSISTANT/ASSOCIATE/FULL PROFESSOR** level in the area of cardiovascular biology. We seek candidates who have already established a highly competitive research program or who will be able to develop such a program.

The Department currently has 24 full-time faculty with interests in cardiac function and disease, blood pressure regulation, ion transport, signal transduction, and structural biology (nuclear magnetic resonance and X-ray crystallography). Core facilities support production of transgenic and knockout mice, gene microarray analysis, proteomics and advanced imaging and microscopy (electron and confocal).

For further information, please see **website: http://www.molgen.uc.edu/logic/.** Applicants should submit curriculum vitae, brief description of research, and the names of three qualified references to: **Jerry B. Lingrel, Ph.D., Professor and Chair, Department of Molecular Genetics, Biochemistry and Microbiology, P.O. Box 670524, Cincinnati, OH 45267-0524.**

DEPARTMENT CHAIR. The Department of Chemistry and Biochemistry at the University of Maryland, Baltimore County (UMBC) invites applications and nominations for the position of Department Chair. The Chair will be expected to provide vigorous leadership for a growing Department of eighteen tenured or tenure-track faculty and four instructors. The Department offers Ph.D. degrees in both chemistry and biochemistry, and applications are invited from internationally recognized scholars in any area of these disciplines who have a commitment to research and quality teaching at both the undergraduate and graduate levels.

UMBC, a Carnegie-ranked research university is a member of the University System of Maryland and is located in a Baltimore suburb about 35 miles from Washington, D.C. To apply send a resume and the names of three persons who can be contacted for supporting letters to: **Dr. M.F. Summers, Department of Chemistry and Biochemistry, 1000 Hilltop Circle, Baltimore, MD 21250.** Applications will be accepted until the position is filled. *UMBC is an Equal Opportunity/Affirmative Action Employer. Minority, women, and individuals with disabilities are encouraged to apply.*

POSITIONS OPEN

UNCW

ASSISTANT PROFESSOR QUANTITATIVE ECOLOGIST

The Department of Biology and Marine Biology at the University of North Carolina, Wilmington (UNCW), invites applications for a tenure-track position starting August 2007. Candidates in any subdiscipline of quantitative ecology/ecological modeling may apply. Duties include undergraduate and graduate teaching, maintaining an active research program, and directing graduate students. The Department offers B.S., M.S., and Ph.D. degrees. Excellent support for research is provided both on campus and at the Center for Marine Science **(websites: http://www.uncw.edu/bio** and **www.uncw.edu/cmsr).** Candidates must have a Ph.D. and postdoctoral experience. To apply, complete the online application process at **website: http://consensus.uncw.edu** by electronically submitting separately (1) a letter of application including brief statements of teaching and research interests, (2) curriculum vitae, and (3) contact information for three references. Microsoft Word or Adobe PDF attachments are preferred. For questions about the position, contact **Dr. Joseph Pawlik, Ecologist Search Chair, e-mail: pawlikj@uncw.edu** or **telephone: 910-962-2377.** For questions about the online application process, contact **Ms. Debbie Cronin, e-mail: cronind@uncw.edu** or **telephone: 910-962-3707.** Screening of applications will begin 20 October 2006. *Under North Carolina law, applications and related materials are confidential personnel documents and not subject to public release. Criminal background checks will be conducted on finalists prior to offers of employment. UNCW is an Equal Opportunity/Affirmative Action Employer. Women and minorities are encouraged to apply.*

ASSISTANT PROFESSOR (TENURE TRACK)
Department of Biology
Morehouse College
Atlanta, Georgia

The Department of Biology at Morehouse College seeks an outstanding early career scientist for a nine-month, tenure-track faculty position at the **ASSISTANT PROFESSOR** level. We seek a broadly trained individual possessing a Ph.D. with postdoctoral training in a biomedical discipline. The candidate must establish and maintain a research program with the potential to incorporate genomics/bioinformatics for data analysis in their research activities.

Morehouse College is a liberal arts institution which is a part of a consortium of schools designated the Atlanta University Center. The successful candidate will be part of a dynamic and well-established life science faculty. Teaching obligations will include introductory level biology. Advisement and mentoring are expected of all faculty members. The proposed starting date is August 2007; salary is commensurate with experience.

To ensure full consideration, applicants must submit curriculum vitae, description of teaching experience and philosophy, description of research interests and career goals, and the names and contact information for three references to: **David B. Cooke, III, Professor and Chair, Department of Biology, Morehouse College, 830 Westview Dr. S.W., Atlanta, GA 30314,** or by **e-mail: dcooke@morehouse.edu.** Deadline is January 1, 2007.

Morehouse College is an Equal Opportunity/Affirmative Action Employer.

MOLECULAR CELL BIOLOGIST/MOLECULAR MICROBIOLGIST. The Department of Biology at Sonoma State University invites applications for this tenure-track position beginning fall 2007. Areas of specialty include, but are not limited to, immunology, medical microbiology, virology, and molecular genetics. See full announcement on our webpage **(websites: http://www.sonoma.edu/biology** or **www.sonoma.edu/aa/fa/tenure_track.shtml).** For additional information, contact **Dr. Dan Crocker, e-mail: crocker@sonoma.edu; telephone: 707-664-2189).** Review of applications will begin October 27, 2006.

POSITIONS OPEN

TENURE-TRACK POSITION
Hendrix College

Applications are being accepted for a faculty appointment beginning August 2007 in the Department of Chemistry. Candidates must be committed to excellence in teaching and biochemical research. Ph.D. required, postdoctoral experience preferred. Competitive startup funding and research time assignment provided in new facilities. The successful candidate will teach biochemistry and participate in general chemistry and nonmajors chemistry courses. While the appointment will likely be at the level of **ASSISTANT PROFESSOR**, applications for other ranks will be considered. Details regarding this position are available at **website: http://www.hendrix.edu/chemsearch.** Application should include a letter addressing the candidate's interest in teaching in a demanding liberal arts environment, a detailed research plan, curriculum vitae, three letters of recommendation (including the telephone numbers and e-mail addresses of the referees), and transcripts of all graduate and undergraduate work. Application materials should be sent to: **Dr. Warfield Teague, Department of Chemistry, Hendrix College, 1600 Washington Avenue, Conway, AR 72032.** Review of the applications will begin on October 16, 2006, and will continue until the position is filled. Hendrix is a distinguished liberal arts college with an endowment of $160 million, sheltering a chapter of Phi Beta Kappa, located in Conway, Arkansas, thirty miles from Little Rock at the foothills of the Ozark Mountains. The College, related to the United Methodist Church, has a strong commitment to excellence in teaching liberal arts. Please visit our **website: http://www.hendrix.edu.** *Hendrix is an Equal Opportunity Employer. Women and members of minority groups are especially encouraged to apply.*

FACULTY POSITION
Structural/Physical Biochemistry

The Institute of Molecular Biology and the Department of Chemistry at the University of Oregon **(website: http://www.molbio.uoregon.edu; www.uoregon.edu/~chem/)** seek applications to fill a tenure-related biochemistry faculty position in the Chemistry Department at the **ASSISTANT, ASSOCIATE, or FULL PROFESSOR** level, as appropriate, to begin in fall 2007 or later. The successful candidate will be expected to teach at the undergraduate and graduate levels, and conduct an aggressive, competitive research program. Individuals studying fundamental problems in cell and molecular biology using structural, biochemical, and/or biophysical approaches are especially encouraged to apply. Interested persons should send curriculum vitae, statement of research plans and teaching interests, and arrange for three letters of recommendation to be sent to: **Biochemistry Search Committee, Department of Chemistry, 1253 University of Oregon, Eugene, OR 97403-1253.** To be assured of full consideration, application materials must be received by November 1, 2006, but the search will remain open until the position is filled. *The University of Oregon is an Equal Opportunity/Affirmative Action Institution committed to cultural diversity and compliance with the Americans with Disabilities Act. Women and minorities are encouraged to apply. We invite applications from qualified candidates who share our commitment to diversity.*

POSTDOCTORAL POSITIONS at Rutgers University to discover, study, and develop botanical therapeutics for diabetes, obesity, and inflammation. Expertise in cell biology/signal transduction, pharmacognosy, or plant biochemistry related to isolation and characterization of bioactive botanicals. Strong publication record and excellent verbal and written communication skills are required. Please submit curriculum vitae, summary of research interests, and the names of three references to: **Ms. Barbara Halpern, c/o Dr. Ilya Raskin, Biotech Center, Rutgers University, 59 Dudley Road, New Brunswick, NJ 08901. E-mail: halpern@aesop.rutgers.edu.** *Rutgers University is an Affirmative Action/Equal Opportunity Employer.*

Faculty Position
Structural Biology

The Department of Biological Sciences at the University of Pittsburgh invites applications for a full-time tenure-track faculty appointment in the area of macromolecular structure and function, pending budgetary approval. We anticipate making this appointment at the **ASSISTANT PROFESSOR** level. Anticipated start date is September 2007. Preference will be given to applicants who conduct problem-oriented research using innovative structural/biophysical approaches and who therefore would welcome an interactive, interdisciplinary departmental environment. The successful candidate will be encouraged to participate in a multidisciplinary graduate program in Molecular Biophysics and Structural Biology that includes faculty from the University of Pittsburgh, the University of Pittsburgh School of Medicine, and Carnegie Mellon University. The successful candidate must have a Ph.D., postdoctoral experience, and will be expected to establish an extramurally funded research program, train graduate students, and participate in undergraduate education.

In order to ensure full consideration, applications must be received by **October 25, 2006**. Applicants should email a single PDF document containing curriculum vitae, a statement of research accomplishments and goals, and a brief description of teaching interests to **biojobs@pitt.edu**. In addition, applicants should arrange to have at least three letters of reference sent to:

Search Committee
Department of Biological Sciences
University of Pittsburgh
Pittsburgh, PA 15260
(412) 624-4266

The University of Pittsburgh is an Affirmative Action, Equal Opportunity Employer. Women and members of minority groups under-represented in academia are especially encouraged to apply.

The University of Texas at Austin

Eukaryotic Molecular Biology Positions
The Institute for Cellular and Molecular Biology

The Institute for Cellular and Molecular Biology, Alan Lambowitz, Director, invites applications for tenure-track/tenured positions in eukaryotic molecular biology. Academic appointments at the level of Assistant, Associate, or Full Professor will be in an appropriate academic unit in the College of Natural Sciences. Candidates should have an outstanding record of research productivity and a research plan that utilizes molecular and biochemical approaches to address important problems in eukaryotic molecular biology. Areas of particular interest include but are not limited to chromatin structure, regulation of gene expression, microRNAs and RNA interference, DNA damage responses, and cell cycle control.

Building on a strong existing faculty, the Institute has recruited more than 40 new faculty members over the past eight years (see www.icmb.utexas.edu). In addition to it's highly interactive and interdisciplinary research environment, the Institute provides administrative and financial support for the Graduate Program in Cell and Molecular Biology and state-of-the-art core facilities including DNA sequencing, mass spectrometry, electron and confocal microscopy, DNA microarrays, robotics, and mouse genetic engineering. A recently instituted MD-PhD program with the UT Medical Branch and the forthcoming Dell Pediatrics Research Institute will further enhance the environment for Biomedical Research.

Austin is located in the Texas hill country and is widely recognized as one of America's most beautiful and livable cities.

Please apply on-line at http://www.icmb.utexas.edu/apply between Sept. 1 and Nov. 1, 2006.

The University of Texas at Austin is an Equal Opportunity Employer. Qualified women and minorities are encouraged to apply; a background check will be conducted on applicant selected.

Cardiovascular Scientist
The University of Montana and the International Heart Institute of Montana Foundation

The UM Department of Biomedical and Pharmaceutical Sciences, in a unique partnership with the International Heart Institute of Montana (IHI) at St. Patrick Hospital and Health Sciences Center, is seeking applications for a cardiovascular scientist to build and maintain a strong extramurally funded research program in one or more of the following areas: cardiovascular pharmacology and toxicology, cardiovascular tissue engineering, in vivo imaging of the cardiovascular system, pathobiology of cardiovascular disease. This is a tenure-track position at the Assistant/Associate/Full professor level with a highly competitive salary and start-up package. The successful candidate will play a major role in bridging the UM and IHI programs. Laboratory space is available at the Hospital and in a new 60,000 sq.ft. research facility at The University of Montana. Biomedical research in the College of Health Professions and Biomedical Sciences is heavily NIH funded and includes emerging strengths in cardiovascular toxicology, protein structure function and tissue engineering. Research strength at the IHI includes valvular heart disease, congestive heart failure, and vascular inflammation. A major federally funded project at the IHI centers on a tissue engineered vascular conduit. Translational research that fosters clinical research and product development is encouraged.

Position requirements include an M.D., Ph.D. or PharmD., a strong record of accomplishment in cardiovascular research, preferably NIH funded; and teaching interests/abilities applicable to graduate programs in biomedical sciences and public health and the professional pharmacy curriculum. Send letter of application, CV, statement of research goals, and contact information for three references to: **Chair, Cardiovascular Search Committee, Department of Biomedical and Pharmaceutical Sciences, SB 270, The University of Montana, Missoula, MT 59812-1552**. For further information contact Dr. J. Douglas Coffin or Dr. Carlos Duran. Screening of applicants will begin **December 1, 2006** and continue until the position is filled.

J. Douglas Coffin, Ph.D.
406-243-4723
Email: douglas.coffin@umontana.edu
Website: www.health.umt.edu/pharmsci

Carlos Duran, M.D., Ph.D.
406-329-5668
timd@ihimontana.org
www.ihimontanafoundation.org

The University of Montana is the recipient of an NSF-ADVANCE award focused on increasing the presence of women in sciences. Equal opportunity/ Affirmative Action/ADA Employer.

EVOLUTIONARY BIOLOGY/ECOLOGY MICROBIOLOGY
The University of Texas at Arlington

The Department of Biology invites applications for two tenure-track positions at the rank of Assistant Professor to complement existing research strengths in ecology, evolution, and genomics. Participation in the Quantitative Biology doctoral program is expected.

- An **evolutionary biologist/ecologist:** Research interests may include but are not limited to population/community ecology, ecological genomics or population/quantitative genetics. **Dr. Laura Gough, Chair of Evolutionary Biology/Ecology Search**
- A **microbiologist:** Research interests may include but are not limited to microbial ecology, virology, genomics, or systematics and evolution. Participation in the undergraduate Microbiology Degree program is expected. **Dr. Thomas Chrzanowski, Chair of Microbiology Search**

Applicants must have a Ph.D. and a demonstrated record of research productivity. Successful candidates will be expected to establish vigorous, extramurally funded research labs and participate in both graduate and undergraduate programs. Located in the Dallas/Fort Worth metropolitan area, UT Arlington is a fast-growing, comprehensive university in The University of Texas System. Additional information is available at **http://www.uta.edu/biology/**. Applicants should submit curriculum vitae; copies of up to five publications; statements of research and teaching interests; and the names, e-mail addresses, and telephone numbers of four persons who can provide letters of reference. Send applications to the appropriate **Search Chair at Department of Biology, University of Texas at Arlington, Box 19498, Arlington, TX 76019-0498**. Review of completed applications will begin **9 October 2006**, and will continue until the positions are filled. Hiring will be contingent on the completion of a satisfactory criminal background investigation for security sensitive positions.

UT Arlington is an Equal Opportunity/Affirmative Action Employer.

POSITIONS OPEN

EVOLUTIONARY BIOLOGIST

Hendrix College Biology Department is seeking an outstanding early career scientist with research expertise in some aspect of evolutionary biology for a tenure-track faculty position at the **ASSISTANT PROFESSOR** level. The successful candidate must have a Ph.D. at time of appointment, the desire to develop an externally funded research program, and most importantly the ability and desire to teach and involve undergraduates in an exciting area of research. Expectations for this position are that 75 percent will be devoted to classroom teaching with 25 percent devoted to student research, with a 50/50 split the first year while initiating the research program. Hendrix will provide startup funds and space for research in the modern D.W. Reynolds facility. Specific courses taught must include at least one core course and an appropriate upper-level course complementary to our current offerings **(see website: http://www.hendrix.edu).**

The proposed starting date will be August 2007, with the possibility of starting in January 2007. Consideration of applications will begin November 1, 2006, and continue until a successful candidate is hired. The Biology Department website **(website: http://www.hendrix.edu/biology/)** will continue to post this ad and provide up-to-date information until the position is filled.

Applicants must submit curriculum vitae, a statement of teaching interest, a statement of research interest, and provide telephone and e-mail contact information of referees as well as arrange for three letters of reference to be sent. Please send all materials electronically to **e-mail: shaw@hendrix.edu,** or hard copies to: **Evolutionary Biologist Position, Dr. Bruce Haggard, Chair, Biology Department, 1600 Washington Avenue, Conway, AR 72032**

Hendrix is a distinguished liberal arts college with an endowment of $160 million, sheltering a chapter of Phi Beta Kappa, located in Conway, Arkansas, 30 miles from Little Rock at the foothills of the Ouachita Mountains. The College, related to the United Methodist Church, has a strong commitment to excellence in teaching liberal arts. Please visit our website at **website: http://www.hendrix.edu.** *Hendrix is an Equal Opportunity Employer. Women and members of minority groups are especially encouraged to apply.*

INTERDISCIPLINARY SCIENTIST
RESEARCH GEOLOGIST/BIOLOGIST
Department of Paleobiology
National Museum of Natural History
Smithsonian Institution

The Smithsonian's National Museum of Natural History seeks a **PALEONTOLOGIST** to conduct an integrative, collections-based research program in pre-Cenozoic marine invertebrates. The successful candidate is expected to utilize modern methods in pursuing a research emphasis in one or more of evolution, paleoecology, morphology, phylogenetics or biogeography. Frequent publication in peer-reviewed journals in specialty areas would be expected, as is curation of appropriate collections, and participation in the scientific community in a manner commensurate with emerging leadership in the area of specialty.

The position is initially a four-year appointment and will be filled at the GS-12 level (salary range is $65,048 to $84,559 per year commensurate with experience.) Reference specific application procedures in actual announcement, see **website: http://www.sihr.si.edu** or contact **Audrey Davis** at **telephone: 202-275-1005.** The announcement will open on September 20, 2006. Applications must be received by October 27, 2006, and must reference announcement number 06RC-6027. This is an interdisciplinary position to be filled by either a Research Geologist or a Research Biologist, depending upon your discipline. All applicants will be notified by e-mail or telephone, when their application is received. *U.S. citizenship is required. The Smithsonian Institution is an Equal Opportunity Employer.*

POSITIONS OPEN

TENURE-TRACK FACULTY POSITION
in Ecotoxicology
Indiana University, Bloomington

Indiana University invites applications for a tenure-track position in ecotoxicology as part of an interdisciplinary environmental science program. The focus of this program is, in part, on the toxic effects of anthropogenic contaminants on ecosystems, including people. It is anticipated that this position will be in the School of Public and Environmental Affairs.

Successful candidates will help develop this environmental science program, maintain an extramurally funded research program, and participate in undergraduate and graduate teaching.

The applicant's expertise is expected to complement existing faculty in ecology, the atmospheric sciences, biogeochemistry, or toxicology in the School of Public and Environmental Affairs and in the College of Arts and Sciences. Appointments are expected to be at the **ASSISTANT PROFESSOR** level, but a senior appointment is possible for an exceptional candidate.

Review of applications will begin on November 1, 2006, and continue until the position is filled. Applications should include curriculum vitae and a statement of research and teaching interests. Submit application materials to:

Dr. Clinton V. Oster, Jr.
Associate Dean of Bloomington Programs
School of Public and Environmental Affairs
Room 300
1315 E. 10th Street
Indiana University
Bloomington, IN 47405-1701

For more information see **website: http://www.iu.edu/~speaweb/about/employment.edu.** *Indiana University is an Equal Opportunity, Affirmative Action Employer, Educator, and Contractor (minorities/females/persons with disabilities), and is strongly committed to achieving excellence through cultural diversity. The University actively encourages applications and nominations of women, persons of color, applicants with disabilities, and members of other underrepresented groups.*

FACULTY POSITION
Department of Molecular Biology
Princeton University

The Department of Molecular Biology at Princeton University invites applications for a tenure-track faculty position at the **ASSISTANT PROFESSOR** level. We are seeking an outstanding investigator in proteomics with special emphasis on mass spectrometry. Ph.D.s or M.D.s with postdoctoral research experience should submit curriculum vitae, a short summary of research interests, and three letters of reference to **website:http://jobs.princeton.edu,** requisition number 0601243. For full consideration applications should be received by December 1, 2006. For additional information about the Department visit our **website: http://www.molbio.princeton.edu.** You may apply online at **website: http://jobs.princeton.edu**, or for general application information and how to self-identify, see **website:http://web.princeton.edu/sites/dof/ApplicantsInfo.htm.** *Princeton University is an Equal Opportunity Employer and complies with applicable EEO and Affirmative Action regulations.*

EXECUTIVE DIRECTOR
Provincetown Center for Coastal Studies

Experienced leader, administrator, and fundraiser with background in marine/environmental field for 501(c)(3) organization engaged in marine research, education, and public policy with 25 full-time staff, 10 seasonal staff, and $2.2 million annual budget. Compensation competitive. See **website: http://www.coastalstudies.org** for description. Letter, resume, compensation requirements, and references to **e-mail: edsearch@coastalstudies.org** by October 31, 2006.

POSITIONS OPEN

PHARMACOLOGY FACULTY POSITION
Touro University, California

Touro University California, located in the northern San Francisco Bay Area, is accepting applications for a full-time, open-rank faculty position in pharmacology. Primary responsibilities include participating in team-taught, integrated basic medical science courses for osteopathic and physician assistant medical students.

Successful candidates will have a strong interest in medical education. Experience in teaching at the professional school level is preferred. Experience in curriculum development would be a plus.

This position emphasizes medical education but also offers the opportunity to do collaborative research or to develop an independent research program.

Review of applications will begin on October 1, 2006, and will continue until the position is filled.

Interested candidates should submit curriculum vitae, statement of teaching and research interests, and contact information for three references to: **Dr. Nathalie Garcia-Russell, Touro University College of Osteopathic Medicine, 1310 Johnson Lane, Vallejo, CA 94592,** or via **e-mail: ngarciarussell@touro.edu.**

POSTDOCTORAL/RESEARCH ASSISTANT PROFESSOR POSITION

Postdoctoral position is available for individual interested in joining an expanding interdisciplinary prostate and urologic cancer program, focusing on the roles of a novel tumor suppressor in prostate cancer. The applicants should have Ph.D. and/or M.D. degree and have experience in pharmacology, structural biology, biochemistry, or *Caenorhabditis elegans* biology. Strong candidate with postdoctoral training may be considered for a Research Assistant Professor position. We offer competitive salary and benefits. Applicants should send curriculum vitae, a letter describing their research interest, and the names of three persons from whom references can be obtained to: **Zhou Wang, Ph.D., University of Pittsburgh School of Medicine, Department of Urology, Shadyside Medical Center, 5200 Centre Avenue, Suite G40, Pittsburgh, PA 15232. Fax: 412-623-3904. E-mail: wangz2@upmc.edu.** *The University of Pittsburgh is an Affirmative Action/Equal Opportunity Employer.*

ASSISTANT PROFESSORSHIP IN CHEMISTRY
Harvard University
Department of Chemistry and Chemical Biology

Applicants are invited to apply for tenure-track Assistant Professorships in all fields of chemistry. Applicants should arrange to have three letters of recommendation sent independently and should provide curriculum vitae, a list of publications, and an outline of their future research plans. Applications and supporting materials should be sent to: **Chair, c/o Ms. Carol Gonzaga, Department of Chemistry and Chemical Biology, Harvard University, 12 Oxford Street, Cambridge, MA 02138-2902.** Reference position: JFCCB133B. The deadline date for receipt of applications and supporting materials is October 15, 2006. *Harvard University is an Affirmative Action, Equal Opportunity Employer. Applications from and nominations of women and minority candidates are strongly encouraged.*

POSTDOCTORAL POSITION
Harvard Medical School

Postdoctoral positions are available to study the mechanisms of translational control in neurons, and the role of dendritic protein synthesis in synaptic plasticity, cognition and neuropsychiatric disease (*Cell* **116:**467, 2004, *Neuron* **44:**59, 2004).

Candidates should be highly motivated and have expertise in molecular biology, biochemistry, or electrophysiology. Please send curriculum vitae and names of three references to: **Dr. Ray Kelleher, Center for Human Genetic Research, Massachusetts General Hospital, 185 Cambridge Street, Boston, MA 02114, e-mail: kelleher@helix.mgh.harvard.edu.**

EUROPEAN SCIENCE FOUNDATION

EUROHORCs

European Young Investigator Awards (EURYI)

Call for Proposals

The European Young Investigator Awards (EURYI) scheme is designed to attract outstanding young scientists, in all research domains including the humanities, from any country in the world to create their own research teams at European research centres.

How EURYI operates

The EURYI Scheme is an exciting opportunity for young researchers, promoted by the Heads of European Research Councils (EuroHORCs) in collaboration with the European Science Foundation (ESF). The aim of the Scheme is to enable outstanding young researchers from all over the world to work in a European environment for the development of European science and humanities and to build the next generation of leading European researchers. EURYI awardees are selected in two steps. The first step consists of application to and selection by the participating organisation from the proposed host country. In the second step ESF selects the awardees from among the national selected candidates using top level, broad disciplinary panels. Awards may be held in any of the fifteen countries participating in this Call: Austria, Czech Republic, Finland, France, Germany, Greece, Hungary, Italy, the Netherlands, Poland, Portugal, Spain, Sweden, Switzerland and Turkey.

Awards 2007

Up to 25 awards will be made in 2007 with a value of between 150 000 euros and 250 000 euros per year for 5 year periods in any research discipline, including the humanities. The selection criteria will be quality of the applicant, of the proposal and of the host institution. EURYI awards are designed to foster original, groundbreaking research. Applicants should have an excellent track record with the potential to become world class leaders in their field of research. Awards will be subject to the terms and conditions of the participating organisation through which the EURYI application is made.

Eligibility

The EURYI Scheme is open to researchers from anywhere in the world who are between 2 and 8 years after receiving their PhD at the closing date of the call, taking into account career breaks.

Further details about the Scheme and the application process are available from the websites of the participating organisations and at: www.esf.org/euryi

Closing Date for Applications: 30 November 2006
The first awards will commence in October 2007.

The work of the European Science Foundation in implementing and coordinating the EURYI Award scheme is supported by funds from the European Commission's Sixth Framework Programme under Contract no. ERAC-CT-2003-510191

For more information about the Call and the EURYI Scheme please contact:

EURYI | ESF | 1 quai Lezay-Marnésia
BP 90015 | 67080 Strasbourg cedex | France
Fax: +33 (0)3 88 37 05 32
E-mail: euryi@esf.org

POSITIONS OPEN

POPULATION GENETICIST, TENURE TRACK

The Department of Biology at California State University, Fresno, is hiring a tenure-track Population Geneticist. The successful candidate is expected to develop a research program that involves both undergraduate and graduate students, to pursue the external funding necessary to maintain a successful research effort, and to teach upper division core courses and undergraduate or graduate courses in their area of specialization. A Ph.D. in genetics or other relevant field is required. Postdoctoral experience is preferred. Send completed application, including form available at **website: http://www.csufresno.edu/aps/vacancy/sc1.pdf,** a cover letter, curriculum vitae, statements of teaching and research philosophy, and three current letters of reference (dated within the last 12 months) to: **Dr. Paul R. Crosbie, Committee Chair, Department of Biology, California State University, Fresno, 2555 E. San Ramon Avenue M/S SB73, Fresno, CA 93740-8034,** or to **e-mail: pcrosbie@csufresno.edu, telephone: 559-278-2074, fax: 559-278-3963.** For full consideration, all materials must be received by 23 October 2006. *California State University, Fresno is an Equal Opportunity Employer.*

ANAEROBIC MICROBIAL PHYSIOLOGY

POSTDOCTORAL POSITIONS available for genome-based studies of the physiology of novel anaerobic microorganisms, with special emphasis on hyperthermophiles and microorganisms capable of dissimilatory metal reduction or reductive dechlorination. Minimum qualifications are a Ph.D. in microbiology or a related field with a research focus in microbiology. Previous experience in the culturing of anaerobic microorganisms and/or systems biology approaches to microbial physiology is highly desirable. For research program details please visit **website:http://www.geobacter.org.** Please e-mail curriculum vitae and names of three references to: **Dr. Derek Lovley, Department of Microbiology, University of Massachusetts, Amherst, MA. E-mail: dlovley@microbio.umass.edu,** or send application materials to: **Search Committee R27006 Microbiology, 203N Morrill IVN, Umass Amherst, MA 01003.** Review of applications will begin on September 12, 2006 and continue until the positions are filled. *University of Massachusetts is an Affirmative Action/Equal Opportunity Employer. Women and members of minority groups are encouraged to apply.*

POSTDOCTORAL POSITION in Epigenetics

The position is at the University of Texas, M.D. Anderson Cancer Center, in the laboratory of **Dr. Mark Bedford.** Research will involve the functional analysis of arginine methylation, and the characterization of proteins that bind methyl-marks on histone tails. You can get some additional information about the Institute, Department, and laboratory at **website: http://sciencepark.mdanderson.org.** Candidates must have a Ph.D. and have published experience in biochemistry and molecular biology. To apply, please send curriculum vitae and three references to **e-mail: mtbedford@mdanderson.org.**

Two **POSTDOCTORAL POSITIONS** are available in the Division of Nephrology at the University of Utah. The research program concerns renal mechanisms of blood pressure regulation and is currently supported by multiple federal grants. Strong background in molecular biology and renal physiology is desirable. Please send curriculum vitae and contact information of three references to: **Margaret A. Amundsen Endowed Professor Dr. Tianxin Yang, Salt Lake City, UT. E-mail: tianxin.yang@hsc.utah.edu; telephone: 801-582-1565, extension 4334.**

MEDICAL PHYSICIST for cancer center in Newark, Delaware. Candidate must have four years of experience and a Master's degree in radiological or medical physics. Must be American Board of Radiology/American Board of Medical Physics-certified or eligible. Fax resume to **P. Bjorklund** at **Christiana Care, fax: 302-325-7047.**

INSTITUT PASTEUR

POSTDOCTORAL FELLOWSHIPS
Institut Pasteur, Paris, France

Founded in 1887 by Louis Pasteur and located in the heart of Paris, the Institut Pasteur is a world-renowned private research organization. The Pasteur Foundation is seeking outstanding Fellowship Applicants. Candidates may apply to any laboratory within 10 departments: cell biology and infection; developmental biology; genomes and genetics; immunology; infection and epidemiology; microbiology; neuroscience; parasitology and mycology; structural biology and chemistry; and virology. See website for details.

Fellowships are $60,000 per year for three years ($45,000 stipend plus $15,000). *U.S. citizenship required.* Deadline: February 2, 2007.

E-mail: pasteurus@aol.com.

Website: http://www.pasteurfoundation.org.

POSTDOCTORAL RESEARCH IN GENETIC ASPECTS OF GLOBAL CHANGE
Department of Biological Sciences
University of Notre Dame

We seek an **EVOLUTIONARY BIOLOGIST** to participate in applied genetic research. This individual will be a founding member of our new Center for Environmental Genomics and will collaborate with **Drs. Hellmann, Lodge, and Feder** on projects involving the impacts of global change. One project examines changes in interspecific hybridization among butterflies under climate change using microsatellites and experimental crosses **(Hellmann).** A second project will develop genetic tools to screen for aquatic, microscopic invasive species **(Lodge and Feder).** A dedicated Technician will support the Postdoctoral Researcher. This is a one-year position with a possible extension of six months; opportunities for grant writing to extend the position are available as is longer-term collaboration. Applicants should send a description of their research, curriculum vitae, and the names of three references to: **Jessica Hellmann, Department of Biological Sciences, 107 Galvin Life Science Center, University of Notre Dame, Notre Dame, IN 46556 (e-mail: hellmann.3@nd.edu).** *The University of Notre Dame is an Equal Opportunity/Affirmative Action Employer.*

Louisiana State University Health Sciences Center (LSUHSC) in New Orleans, Louisiana, has an immediate opening for a **POSTDOCTORAL RESEARCHER.** The overall duties of the researcher in this position are to plan, organize, and conduct highly independent research on two projects: (1) understanding the role of Nischarin in breast cancer cell migration and invasion; and (2) making Nischarin knockout mouse. For further information see **website: http://www.medschool.lsuhsc.edu/biochemistry/faculty_detail.asp?id=1121.**

Applications are sought from highly motivated individuals with strong background in molecular and cell biology. Candidates must have a Ph.D. in biochemistry, molecular or cellular biology, or health-related field. Desire demonstrated experience in molecular techniques such as gene cloning, gene expression, and mammalian cell transfections. Experience with animals is a plus. Interested candidates should send curriculum vitae, a summary of research statement and names, telephone numbers, and e-mail addresses of three references to**: Dr. S. K. Alahari, Department of Biochemistry and Molecular Biology, Louisiana State University Health Science Center, CSRB, New Orleans, LA 70119. E-mail: salaha@lsuhsc.edu.** *LSUHSC is an Affirmative Action/Equal Opportunity Employer.*

POSTDOCTORAL ASSOCIATE
Computational Systems Biology

The laboratory of **Vincent VanBuren, Ph.D.,** part of the newly established Department of Systems Biology and Translational Medicine in the College of Medicine, seeks a Postdoctoral Associate in the broadly defined area of computational systems biology to work in one or more of the following areas: development of novel analyses of microarray data or biological sequence information, reconstruction of biological networks from high-throughput data, or molecular-level simulations of biological systems. Prospective candidates must hold a Ph.D. in a related field, have skills in computer programming and mathematics, have expert knowledge of molecular biology, and must effectively interact with a diverse population of students, faculty, and staff. Additionally, candidates with skills in one or more of the following specific areas will be favorably considered: MATLAB, Perl, Genespring, structural equation modeling, Bayesian methods, DNA microarray analysis, linear algebra, differential equations, data mining, and database management. Interested candidates should send a letter of interest, curriculum vitae, and contact information for three references to **e-mail: vanburen@tamu.edu,** or by regular mail to: **Dr. Vincent VanBuren, Director, SBTM Microarray Laboratory, Assistant Professor, Department of Systems Biology and Translational Medicine, Texas A&M Health Science Center, 702 S.W. H.K. Dodgen Loop, Temple, TX 76504.** Additional keywords: bioinformatics, computational biology, gene regulatory networks, postdoctoral fellow.

Texas A&M Health Science Center is an Equal Opportunity Employer.

FACULTY POSITION, ASSISTANT/ ASSOCIATE OF BIOLOGY, GENOMICS

The Department of Biology at the University of Rochester is continuing its hiring initiative. It is our goal to continue to build a research and graduate education environment that encourages interactions across biological disciplines and integrative approaches to biology. This year we intend to fill a tenure-track position at either the Assistant or Associate level in genomics. Highly qualified candidates in all areas, including functional and comparative genomics, are encouraged to apply. Our research and graduate programs are integrated into a larger research campus, which includes computer sciences, brain and cognitive sciences, biomedical engineering, and the School of Medicine and Dentistry.

Please send curriculum vitae, a statement of research interests, three letters of reference, and one to two reprints to: **Faculty Search Committee, Department of Biology, University of Rochester, Rochester, NY 14627-0211.** Applications may also be submitted as PDF files to **e-mail:ynkr@mail.rochester.edu.** Review of applications will begin November 1, 2006. *The University of Rochester is an Equal Opportunity Employer.*

MARKETPLACE

PICTURE YOURSELF AS A AAAS SCIENCE & TECHNOLOGY POLICY FELLOW!

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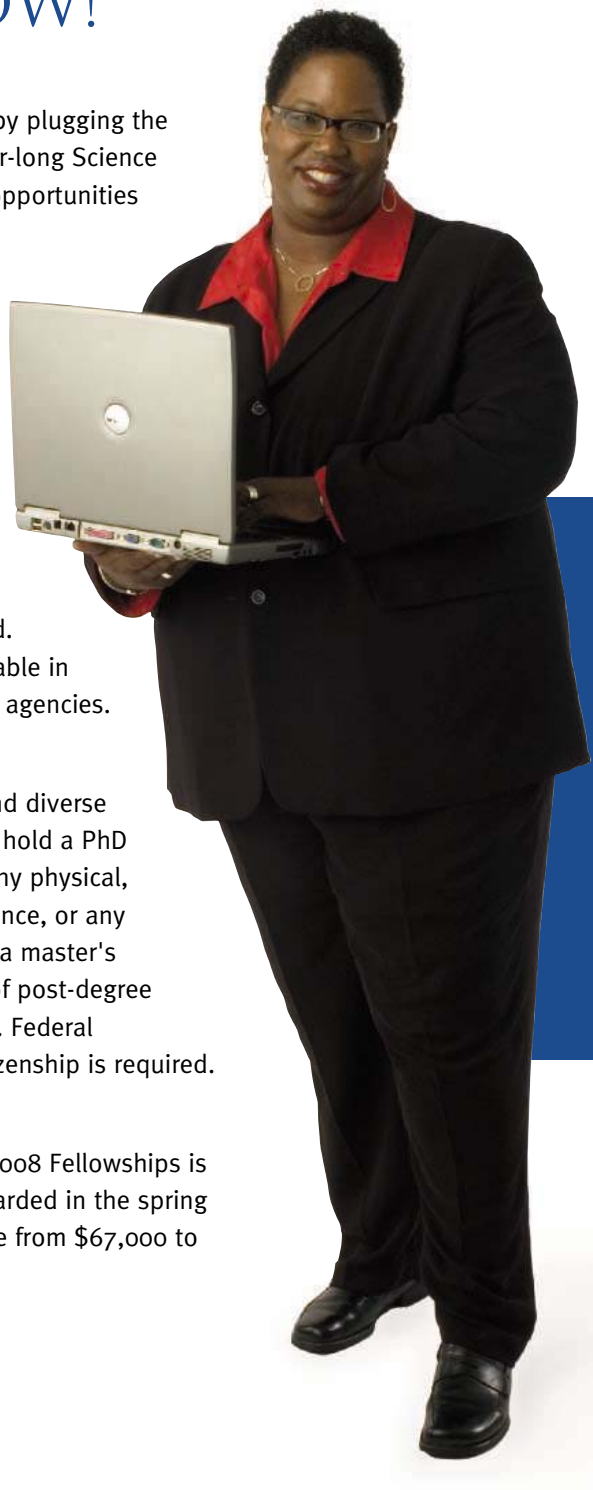
Apply Now!
The application deadline for the 2007-2008 Fellowships is 20 December 2006. Fellowships are awarded in the spring and begin in September. Stipends range from $67,000 to $87,000, depending on experience.
To apply: fellowships.aaas.org



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Stephanie Adams, PhD

Interdisciplinary Engineering, Texas A&M University.

2005-2006 AAAS Fellow at the National Science Foundation, Division of Engineering Education Centers.

Currently Associate Professor and Assistant Dean for Research at the Department of Engineering at University of Nebraska-Lincoln, which granted a two-year Interpersonal Agreement (IPA) for the AAAS Fellowship.

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